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Competing with insects for food, fibre and shelter

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Humans need food, fibre and shelter to survive. To get these, we compete with insects, the most successful group of animals on the planet. We attempt to 'control' the impact of such pest species, often with mixed success. In the past, we've relied on the use of single-strategy approaches. These often fail, either through a poor understanding of the insect's biology and ecology, and/or through changes to the insect's biochemistry or behaviour. Modern pest management rests on a paradigm of Integrated Pest Management (IPM), where a combination of strategies is used to manage pest populations so that their economic impact is minimised. The development of these strategies rest on the talents of many disciplines, from chemistry to biology to economics to product marketing. In developing these systems, the questions have changed from 'How do we kill this pest?' to 'Why do populations of this pest reach levels high enough to cause economic damage?'; this is where ecology plays a vital role. However, there are strong social aspects to the development and adoption of this integrated approach. Development of new products and processes needs to be coordinated and managed. End users (farmers) need to be convinced that adopting these strategies will benefit them; this requires their involvement in the research process. Differences in pest biologies and ecologies and in the expectations of end users mean that there is no simple recipe for successful IPM. Creativity and innovation are vital to feed, clothe and house people.

What's in a Name and Who's your Relative

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Unambiguous communication about our friends and relatives depends upon their having distinctive names. These tell us much about their identity and perhaps something about relationships. Likewise species of organisms, which are the units of biodiversity need unique names. Insects are by far the most diverse group of organisms, with numbers of species easily exceeding 1.5 million. Although many have been named, the completion of the task remains distant, as does discovery of the true diversity in the face of loss of natural habitats and of taxonomists (those that name and classify life). However it is incumbent on us to document who we share our planet with, and to understand the evolutionary relationships amongst them. These are two distinct tasks but the practices and expertise involved in each are complementary. New approaches to both tasks, together with a revival of interest in the taxonomic framework for biodiversity studies, allow some optimism for greater productivity. For example, the U.S. National Science Foundation ring-fenced funds a decade ago to support the transfer of information from the older generation of taxonomists to the younger. The broadly trained output of this PEET (Partnerships for Enhancing Expertise in Taxonomy) program are dispersing and forming centres of expertise, often in non-traditional institutions and in the countries with high diversity that have lacked modern skills. Also emanating from the USA is a mega-project to document the 'Tree of Life' – an ambitious attempt to identify the relationships of all the organisms on the planet, especially using DNA data and bioinformatics tools. Thus far, funding has included provision for study of selected major insect groups – which after all comprise so much of the 'tree'. Australia has the mechanisms in place for comparable efforts, but funding to the ABRS (Australian Biological Resources Study) has been too slight to reverse an ongoing decline in support for taxonomy. Europe especially ought to follow the US model. Major advances in working methods for decribing our biota have resulted from the availability of the internet to disseminate information and the power of computing to manage the extensive databases that are generated by any study of biodiversity. Data management packages complement software designed to help non-experts identify organisms – many of these tools emanated in Australia, some in Brisbane at the University of Queensland. This combination of a newly-recruited skilled cohort of taxonomists with major research tools and appropriate federal support allows us to believe that the task of documenting and understanding those we share our planet with is attainable.

Entomology in Australia – a proud heritage

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Entomology is important to Australia. Insects, and their relatives, have major economic impact, they are vital in the ecosystems and they make great model biological systems. They are also fun. Australian entomologists engage in a wide range of occupations in agriculture, resource management, academic research, biosecurity and in urban systems, to mention a few.
CSIRO Entomology is the oldest Division in CSIRO and throughout its 77 year history CSIRO scientists have worked with others to bring about miracles – control of major pests and weeds, and the documenting our unique biodiversity.

ENVIRONMENTAL CHANGE, MOSQUITOES AND DISEASE: AT WHAT RISK AUSTRALIA?

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Summary: In the last couple of decades, the world has seen the 'emergence' of a whole variety of new diseases, like Ebola fever, HIV/AIDS and SARS. Many familiar diseases have also extended over wider areas and now affect many more people than ever before, including diseases transmitted by mosquitoes (like malaria and dengue fever). Many researchers argue that this is a result of humans causing major changes to the natural environment, thereby creating new opportunities for microbes to infect large numbers of people. Australia is no exception to the many countries with dramatically changed ecosystems, with major modifications that include agriculture, irrigation, mining, deforestation, and urban development. Have these environmental changes left us vulnerable to worsening levels of mosquito transmitted disease? This talk examines the evidence for a relationship between environmental change and the risk of mosquito transmitted disease in Australia, and discusses new threats to our shores from microbes like Japanese Encephalitis Virus.

Use of microarrays to detect insecticide resistance mechanisms in *Anopheles* malaria vectors

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Insecticide resistance is a serious threat to sustainable malaria control in Africa. We have developed a small-scale microarray for the malaria mosquito *Anopheles gambiae*. This array contains unique fragments from all the major genes associated with metabolic resistance to insecticides. These include the cytochrome P450s, esterases, glutathione transferases, partners of the P450 oxidative metabolic complex, redox genes and a large number of control genes, to facilitate normalization and interpretation of the data. We have used this 'detoxification chip' to identify the genes up-regulated in insecticide resistance strains of A. gambiae and the preliminary results of these studies will be presented. In addition, we are also using this chip to address several questions related to the regulation and induction of these supergene families. Finally, the feasibility of using the 'detox chip' to monitor resistance mechanisms in field populations will be addressed.

Effect of Conventional Insecticides on Sunn Pest Egg Parasitoids *Trissolcus grandis* and *T. semistriatus* (Hymenoptera: Scelionidae)

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Lethal and sublethal effects of fenitrothion and deltamethrin, commonly used insecticides for controlling sunn pest *Eurygaster integriceps* Put. in Iran, were assessed on egg parasitoids *Trissolcus grandis* Thoms. and *T. semistriatus* Nees. Adult parasitoids were exposed to fresh residues of the insecticides and LC_{50} bioassays were carried out. LC_{50}s of fenitrothion for *T. grandis* and *T. semistriatus* were 16.2 and 7.9 ppm, respectively. The LC_{50} values of deltamethrin were 155 and 145.6 ppm for *T. grandis* and *T. semistriatus* respectively. Sublethal effects of the insecticides on life table parameters were measured for parasitoids exposed to rates near to LC_{50}'s of the insecticides within the first 24 hrs after emergence. Fenitrothion and deltamethrin reduced longevity by 12.7 and 17.6% for *T. grandis* and 13.2 and 22.5% for *T. semistriatus*, respectively. Reproductive capacity of exposed females was significantly affected in either species. The sex ratio of the progeny of both parasitoids was not affected significantly but was male based. The intrinsic rate of increase for *T. grandis* were 0.298, 0.289 and 0.296 and for *T. semistriatus* were 0.297, 0.293 and 0.295 female offspring per female per day in control, fenitrothion and deltamethrin respectively. Doubling time values for *T. grandis* in control, fenitrothion and deltamethrin were 2.32, 2.40 and 2.40 respectively and for *T. semistriatus* were 2.33, 2.37 and 2.35, respectively. The sublethal effects in this study indicated that sublethal effects must be considered to evaluate accurately the selectivity of insecticides in favor of the parasitoids.

Nectar feeding moths learn single odours during flower visiting.

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Floral odours play a major role in plant-insect communication. We examined the influence of a number of single floral odours on the innate and learnt foraging behaviours of the moth *Helicoverpa armigera*. In wind tunnel and flight cage tests, we showed (1) moths could distinguish between flowers augmented by single odours (2) moths could learn single odours present in flowers on which they had foraging experience, which occurred regardless of whether the flower had a relatively simple or complex odour blend and (3) moths showed the ability to learn more than one component from a single flower, with equal preference for the learnt odours. The results imply that moths can learn single odours within complex floral blends, which may enable them to discriminate among odour profiles of individual flowers from the same species. We discuss this behaviour within the context of nectar foraging in moths and odour signalling by flowering plants.

How do mangrove ants in North Australia survive inundation?

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More than 20 ant species inhabit mangroves in Darwin Harbour, NT, where the tidal movement is up to 8 metres. Some of these species live exclusively in mangroves and have adapted specific strategies to survive the frequent inundations. The most marine living ant species is *Polyrhachis sokolova*, which nest in the mud in places where they can be flooded by 62 % of the high tides and with up to 2 metres of seawater over the nest entrance. During inundation the ants cluster in air-pockets in the nest. The CO_2 concentration in the deeper part of the nest can in some circumstances exceed 10 %. The tide and the movements of the 'ground water' flush the air in the nest, so the CO_2 rich air is replaced with atmospheric air every two weeks. The ant *Camponotus anderseni,* which exclusively nest inside twigs of *Sonneratia alba* trees, prevent intrusion of water into the nest cavities by having a soldier plug its head into the nest entrance. During longer inundation, the CO_2 concentration in the nest chambers can exceed 30%. Simultaneously, the ants reduce their respiratory rates down to 1% of normal respiration. The extreme environmental conditions in the nests of mangrove ants allow investigations of adaptations of the ants to stressful conditions.

The sex pheromone of the first Australian Lymantriidae, *Teia anartoides*: Theromo-labile diverse pheromone blend

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The Painted Apple moth, Teia anartoides (Walker) has been recently introduced to New Zealand. This invasive species has the potential for significant economic and ecological damage to New Zealand horticulture and forestry due to its wide host range. As part of the eradication program initiated by the New Zealand Ministry of Agriculture and Forestry (MAF), the sex pheromone of the Painted Apple moth (the first Australian Lymantriid) was investigated using GC-EAD analysis; female extracts dervatization followed by GC-MS and TLC analysis. The major sex pheromone compound of female Painted Apple moth was identified as 6Z, 9E-heneicosadien-11-one. Other minor pheromone compounds includ: 6Z, 9Z-Heneicosadiene, 6Z-9R,10R-Epoxyeicosene, 6Z-9R,10S-Epoxyheneicosene, 6Z, 9Z-Heneicosadiene-11-ol, 6Z-Heneicosene-11-one and 6Z, 8E-Heneicosadiene-11-one were also identified in the female sex pheromone gland. In field cage experiments, a blend of 6Z, 9E-heneicosadien-11-one (100), 6Z, 9Z-Heneicosadiene (100), 6Z-9R, 10R-Epoxyeicosene (5), 6Z-9R, 10S-Epoxyheneicosene (10), 6Z, 9Z-Heneicosadiene-11-ol (5), 6Z-Heneicosene-11-one (1) and 6Z, 8E-Heneicosadiene-11-one (25) was as attractive as calling females or gland extracts. Field trapping experiments conducted in Auckland, New Zealand supported these results. 6Z, 9E-heneicosadien-11-one was found to be a thermo labile compound and rearranged to other products under ambient temperature, rendering the synthetic pheromone lure inactive after two days. The newly identified pheromone can by used in population monitoring, detection and any eradication program of this invasive species.

Exploiting biodiversity and chemical ecology in a 'push-pull' strategy for management of stemborers in Africa

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Lepidopteran stemborers cause major yield loss in maize production throughout Africa. Studies are described that have led to the development of a 'push-pull' strategy for minimising stemborer damage in maize-based farming systems for eastern and southern Africa. Development of the strategy originally involved the selection of plants that could be employed as trap crops to attract stemborer colonisation away from maize, or as intercrops to repel the pests. The two main trap plants that were identified are Napier grass, *Pennisetum purpureum,* and Sudan grass, *Sorghum vulgare sudanensis,* both of which attract greater oviposition by stemborers than cultivated maize. The intercrops giving maximum repellent effect are molasses grass, *Melinis minutiflora,* and a legume species, *Desmodium uncinatum.* 'Push-pull' trials in farmers' fields reduce stemborer attack and also increase levels of parasitism of stemborers on maize plants, resulting in a significant increase in maize yields. The trap crop and intercrop plants also provide valuable forage for cattle. The plant chemistry responsible for stemborer control involves release of attractant semiochemicals from the trap plants and repellent semiochemicals from the intercrops. In fields intercropped with *M. minutiflora,* parasitism of stemborers is also increased by certain chemicals repellent to ovipositing adults being the same as those produced by maize plants heavily attacked by stemborers.

Lethal and sublethal effects of different ecotypes of *Melia* against *Plutella xyostella* Linn.

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Extracts from drupes of *Melia azedarach* (Linn.) ecotypes growing in three states of India viz. Rajasthan, Punjab and Himachal Pradesh representing different geographical regions i.e. plains, arid and hilly areas, respectively, were evaluated against second instar larvae of diamondback moth, *Plutella xylostella* (Linn.). LC_{50} of the extract from arid region of Rajasthan was the lowest (1.472 %) followed by that from plains of Punjab (1.794 %) and the maximum (2.296 %) for the extract from hilly areas of Himachal Pradesh. The survivors demonstrated various kinds of sublethal effects such as prolongation of larval period, reduction in pupal weight, deformation of pupae (up to 69.68 %) and inhibition of adult emergence (up to 92 %). A highly pronounced curtailment of fecundity (up to 8.5 against 68.5 in control) and significant reduction in oviposition period of the adults (up to 3.5 days against 7.4 in control) was also recorded. The extract from arid region of Rajasthan, with relatively higher mean temperature (28^0 C), low humidity (56 %) and minimum rainfall (15.5 mm), excelled in producing lethal and sublethal effects; only 1.0 % normal adults survived at the end in comparison to 3.5 and 5.5 % in case with the extracts from the plains and hilly areas, respectively. Inferences of the study give important indications for entomologists and tree breeders.

Control of *Liriomyza trifolii* pupae with solar radiation

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Subterranean *Liriomyza trifolii* pupae were successfully controlled with solar radiation after cropping of a leaf vegetable, *Chrysanthemum coronarium* var. *spatiosum*. Covering ground with transparent polyvinylchloride film elevated temperature to 54 C at a depth of 2 cm beneath the soil surface, and no flies emerged to attack the next crop in late May. Lethal high temperature and duration combinations for the pupae were investigated in laboratory, and the pupae were all killed by exposure to either 44 C for 24 h, 46 C for 3 h or 48-54 C for 30 min. Soil temperatures under the film were measured in a closed greenhouse, and maximum high temperatures of 48 C or higher were attained at a depth of 2 cm on a fine day from early April to mid October. Thus the physical control method is effective for practical use from early April to mid October in Osaka prefecture.

Nutritional effects of five different oilseed rape cultivars on the survival rate and fecundity of cabbage aphid *Brevicoryne brassicae* (Homoptera: Aphididae)

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The cabbage aphid, regarded as one of the most important insect pests of oilseed plants in most parts of Iran, causes economic losses on the crop. In the present study, the biological responses of the cabbage aphid fed on 5 rapeseed cultivars namely Urica, Hayola 401, Hayola 308, Parkland and Pf (7045, 91) was investigated at 4-6 phenological leaf stage, under glasshouse conditions of 24^{o}C±1, with 65-75% R. H. and photoperiod of 16:8 (L: D) periods. Antibiosis was determined by studying the percentage survival of the nymphs, duration of their development time, fecundity (total number of progeny per female produced within the first 10 and 15 days reproductive cycle) and finally calculating the relevant intrinsic rate of natural increase (r_m values). The analysis of variance indicated that, there were significant differences ($p<0.05$) between the varieties. At 4-6 leaf stage, the highest mean "r_m" value (0.3229 ± 7.516E-0.3 and 0.3210 ± 7.783E -0.3) was estimated for rearing on Hayola 401 with the smallest (0.1958 ± 1.645 E-0.2 and 0.1957 ± 1.461 E-0.2 on the Pf (7045, 91) within the first 10 and 15 day periods respectively. Thus in this study, Hayola 401 is regarded as the most susceptible and the Pf (7045, 91) as the most resistant variety in relation to the aphid.

Above- and belowground nutritional ecology of the invasive maize pest *Diabrotica virgifera virgifera* (Crysomelidae, Galerucinae) in Europe.

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500 years after the introduction of maize into Europe its most important pest *Diabrotica virgifera virgifera* LeConte (Coleoptera, Chrysomelidae, Galerucinae) has finally caught up. First detected in Europe 1992 near Belgrade it has spread into 15 European countries since then. We studied the nutritional ecology of adults and the larval performance on European maize cultivars and several alternative host plants. 1) The nutritional ecology of adults in Hungary was studied by performing gut content and pollen analyses. Adult beetles were using the majority of all flowering weeds as alternative pollen sources. The use of maize tissue and alternative pollen resources was depending on maize phenology, diversity of flowering weeds and sex of the beetles. 2) We developed a new method, which allowed measuring the food conversion efficiency of *D. v. virgifera* larvae. Larval performance differed significantly between the tested maize varieties. They performed as efficiently on several other grass species (common weeds and winter wheat). The performance was related to the total nitrogen content, the C/N ratio and the phytosterol composition of the host-plants. The results of our study have multiple implications for management-strategies such as crop rotation or weed control and will help to develop IPM strategies for Europe.

The Impact of Areawide Pest Management on Carabids in Indiana/Illinois, USA

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The study was conducted over a 6-year period in a 41.4 km^2 contiguous area. The target pest species was the western corn rootworm (WCR), *Diabrotica virgifera virgifera* LeConte. The study was designed to test the feasibility of using semiochemical (cucurbitacins) insecticide-baits as the primary management tool for WCR on an areawide basis. Observations in 2000 indicated that the insecticide-bait might be negatively impacting some non-target organisms. Therefore, a study was undertaken in 2001-2002 to determine the impact on non-targets. The study was carried out in 8 fields (4 within the WCR managed area and 4 within the control area). Four sampling methods were utilized to gauge impact on non-targets; pitfall traps, sweep-net sampling, Pherocon® AM traps, and direct counts on plants. In the pitfall trapping study, carabids were the most abundant non-target species observed. The data showed that significantly lower numbers of carabids were found within the managed area following treatment when compared to the control area. This indicates that the areawide program had a negative effect on this non-target. However, heavy rainfall confounded the results. It is not known at this point whether the toxic effect was the result of the semiochemical alone, the combination of the semiochemical with the insecticide, the insecticide alone, or one of the previous in combination with rainfall.

Action and regulation of defensin in *Ornithodoros moubata* (Acari: Argasidae).

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Generally, arthropod antibacterial peptides are inducible in response to injury or bacterial infection. On the other hand, *Ornithodoros moubata* shows up-regulated gene expression of defensin in response to blood feeding. In the present study synthetic tick defensin was used to investigate the spectrum and mode of action of natural tick defensin. Although originally purified as a *S. aureus*-killing antibacterial peptide, synthetic tick defensin shows a broad spectrum that kills many other Gram-positive bacteria including medically important *E. faecalis*, *S.aureus* and MRSA at low concentrations. Synthetic tick defensin shows extremely low hemolytic activity against human erythrocytes. The mode of action of defensin was confirmed to be due to the peptides interacting with and disrupting the cytoplasmic membrane. Tick defensin seems to also interfere with cell separation as shown by the presence of multiple aberrant cross-walls. This is the first report of abnormal cell division as a result of defensin treatment. *Ornithodoros* defensin gene expression appears to be mediated by a newly cloned Rel/NF-κB family transcription factor OmRel. Synthetic tick defensin demonstrates high antibacterial activity against Gram-positive bacteria and low hemolytic activity suitable for pharmaceutical applications.

Biosynthesis of worker mandibular gland components in social bees.

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Biosynthetic pathways of several worker mandibular components in *Apis cerana japonica* and *A. mellifera* were examined by incorporation experiments using deuterate labelled compounds.

Molecular Cloning and Recombinant Expression of Antimicrobial Peptides from House Fly, *Musca domestica*

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Two antimicrobial peptide genes, the cecropin and defensin, were cloned by from the houseflies, *Musca domestics*. The full length of the cecropin cDNA was 403 bp and the open reading frame of the cDNA encoded a 63 amino acide procecropin. A potential signal sequence of twenty-three amino acids was identified, ending with the residues QSEA. A single cleavage reaction at Ala-23 therefore would generate the mature forty-amino acid *Md*-Cec peptide. The N-terminal portion of the molecule is highly basic, while the C-terminal domain is predominantly hydrophobic. The sequence identity with other cecropin was between 80% and 97%. The open reading frame of the defensin encoded a 92-amino acid peptide, which contained an NH_2-terminal signal sequence (1~22) followed by a propeptide and the mature peptide (53~92). The sequence identity with other insect defensin is between 51% and 73%. The mature peptide, with a predicted molecular weight of 4.0 kDa, and pI of 8.69, has 1 negative charged amino acid and 4 positive ones. The cecropin and defensin were expressed in *E. coli* and yeast and the recombinant peptides in yeast had the activity against several Grame negative and positive bacteria. The recombinant cecropin and defensin were confirmed by amino acid sequencing.

RAPD molecular marker identification in NB_4 D_2 and NB_{18} Silk worm *(Bombyx mori)*

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Bivoltine races of silk worm *(Bombyx mori)* NB_4D_2 and NB_{18} are subjected to genomic DNA isolation and purification. The purification of DNA is subjected to PCR amplification to know the polymorphic and monomorphic RAPD molecular marker through Agrose gel electrophorosis method. The number of polymorphic and monomorphic bands indicated degree of divergent in silk worm races. After knowing the sequence of DNA amplification also recorded the significance among the races to adopt the various geo-eco physiological conditions by knowing molecular marker sequence which are target at the level of DNA. Further degree of genetic relationship will be discussed in detail.

Molecular marker of genomic DNA in Silk worm (Bombyx mori)

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The genomic DNA of silk worm *(Bombyx mori*) one of the race called NB_4 D_2 extracted and purified as per the standard method. Silk worm genomic library of 0.5 - 2.0 kb in length was generated by digesting NB_4 D_2 genomic DNA with Pst_1 restriction endo nucleus and is ligated Pst_1 digested dephosphorylated PUC_{18} plasmid. The recombinant clones were selected by dot blotted and hybridized with translated radio labeled NB_4 D_2 genomic DNA. 248 out of 1055 recombinant plasmid which carrying only single insert and showing absence of hybridization signal selected as a probes for RFLP separation.

Genetic differentiation in three Cryptophlebia **(Lepidoptera: Tortricidae)** *species of economic importance in South Africa*

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The false codling moth, *Cryptophlebia leucotreta,* is one of the most damaging and catholic insect pests in South Africa. The pattern of genetic differentiation was studied in this species and compared with those of the litchi moth *C. peltastica* and the macadamia nut borer *C. batrachopa*. Populations of all three species were collected throughout their known range in South Africa from a variety of tropical, subtropical and deciduous hosts. Populations were studied using amplified fragment length polymorphism (AFLP) analysis and nucleotide sequence variation of the mitochondrial cytochrome oxidase 1 (COI) gene. Significant intraspecific variation was detected in all three species although *C. leucotreta* appeared as the most variable species, while *C. batrachopa* was the least variable. Analysis of molecular variance (AMOVA) revealed a high level of variation within populations for all three species. Estimates of the absolute number of migrants between populations (Nm) based on data obtained from mitochondrial and nuclear DNA markers indicate that *C. leucotreta* has a strong geographical structure with low gene flow between geographic populations. However, populations collected from different hosts differed only slightly. Furthermore, evidence was found for gender related dispersal. The results obtained are explained and used to provide insight into managing population levels of *Crytophlebia* spp. in different environments.

Assessment of bacteriophage WO involvement in *Wolbachia*-induced CI within the *Culex pipiens* complex

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The *Culex pipiens* complex includes some of the most medically important mosquitoes due to their cosmopolitan distribution and their capacity to vector infectious diseases. Members of the complex differ in their ecology, behavior, physiology and vector competence. To date, no consensus has been achieved about the taxonomical status of representatives of this group. Opinions of their relationship range from distinct species with no gene flow to physiological forms with considerable introgression. Most populations of *Culex pipiens* are infected with *Wolbachia*, a maternally inherited bacterium, which causes cytoplasmic incompatibility (CI) resulting in embryonic mortality. In prior molecular characterization, we have demonstrated that *Culex* populations differ in the bacteriophage WO that is thought to infect *Wolbachia* (Sanogo and Dobson, 2004). To test the hypothesized involvement of bacteriophage WO in determining CI in *Culex*, crosses have been conducted between nine *Culex* strains that differ in their bacteriophage WO infection types. Here, we will present the crossing results and discuss the correlation between bacteriophage WO and CI type. We will also discuss the results in relation to the restriction of gene flow among naturally occurring *Culex pipiens* populations. Reference: Sanogo YO and SL Dobson. 2004. Insect Molecular Biology (*in Press*)

Japanese oak wilt as a newly emerged forest pest in Japan: why does a symbiotic ambrosia fungus kill host trees?

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"Japanese oak wilt (JOW) has been known since the 1930s, but in the last fifteen years epidemics have intensified and spread to the islands' western coastal areas. The symbiotic ambrosia fungus *Raffaelea quercivora* is the causal agent of JOW, and is vectored by *Platypus quercivorus*. This is the first example of an ambrosia beetle fungus that kills vigorous trees. Mortality of *Quercus crispula* was approximately 40% but much lower for associated species of Fagaceae, even though each species had a similar number of beetle attacks. It is likely that other oaks resistant to the fungus evolved under a stable relationship between the tree, fungus and beetle during a long evolutionary process. *Quercus crispula* was probably not part of this coevolution. *Platypus quercivorus* showed the least preference for *Q. crispula*, yet exhibited highest reproductive success in this species. Therefore, *P. quercivorus* could spread more rapidly in stands with a high composition of *Q. crispula*. *Platypus quercivorus* can reproduce only one year on each *Q. crispula* tree. The relationship among *Q. crispula – R. quercivora – P. quercivorus* seems evolutionary unstable. JOW seems to be an invasive pest of *Q. crispula*. Concentric patterns of JOW spread also support this hypothesis.
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A new approach To field application of sterile insect technique (Sit) in the management of red palm weevil, *Rhynchophorus Ferrugineus* Oliv. (Coleoptera: Curculionidae).

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The red palm weevil *Rhynchophorus ferrugineus* Oliv. is one of the most devastating pests of palms especially coconut all over the world. Being an internal borer the infestation by this pest will be visible only at the later stage when the palm collapses. Efforts were taken to manage the red palm weevil in the coconut garden using sterile insect technique. A new approach to this technique was devised by studies at the Department of Entomology, College of Agriculture, Vellayani, Thiruvananthapuram, Kerala, India. In this new method the initial population was estimated by relative method by counting the different life stages of the weevil present inside the infested palms. The male weevils were sterilized using Co-60 gamma irradiation source and released in the ratio of 10:1 of sterile to normal. Weevils were then recaptured using pheromone traps and were assessed for sterility. The life table studies revealed that the multiplication rate of red palm weevil as eight and based on that subsequent generation releases were carried out. The overall studies showed that release of sterile insects at 15 days interval for 17 times in three generation was sufficient to manage the red palm weevil effectively at field level.

The effect of kaolin particle films on pests of field vegetable crops.

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The revocation of many commercial insecticides has increased the need to evaluate practical alternatives for the control of insect pests on field vegetable crops. One innovative approach being pursued is the use of novel physical barriers that can be applied to crops to protect them from insect attack. The use of kaolin particle films that are sprayed onto the plants has previously been assessed and used successfully in the protection of top fruit. Our aim is to investigate the potential of these particle films for pest management in field vegetable crops. The studies include both field trials and laboratory investigations, the latter concentrating on particular pests of the Brassicaceae including *Delia radicum*, *Myzus persicae*, *Plutella xylostella* and *Pieris brassicae*. Results from the laboratory studies and preliminary field trials will be presented.

Resistance cultivars & species of the woody ornamental *Lagerstroemia* spp. to the flea beetle, *Altica litigate*.

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"The woody ornamental crape myrtle (*Lagerstroemia* spp.) is a most popular flowering shrub/small tree due to its colorful and lengthy summer flowering and a relatively low landscape maintenance that includes an almost pest-free nature. *Altica litigate*, a metallic blue-green flea beetle that develops high populations on several species of native wildflower primroses (*Oenothera* spp.), can severely damage young crape myrtle plants in container production. In a field (open nursery) study, containerized liners of eleven cultivars of crape myrtles, representing two species (*L. indica* and *L. fauriei*) and their interspecific hybrids, were exposed to high populations of flea beetles to observe their feeding choice and damage. In laboratory experiments young leaves from these crape myrtle cultivars were used to bioassay the flea beetle adults in a no-choice feeding study. Cultivars of *L. indica* were the most susceptible to have severe damage and defoliation by flea beetles, while a remarkable resistance was found in *L. fauriei* cultivars and most of its interspecific hybrids.
"

Insecticidal activity of extracts from Australian flora.

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Extracts from approximately 450 different indigenous plant species in the families Lamiaceae, Myrtaceae and Proteaceae were screened in the laboratory for their bioactivity against a number of key arthropod pests, including two-spotted mite (TSM), Tetranychus urticae, greenhouse thrips (GHT), Heliothrips haemorrhoidalis, and diamondback moth (DBM), Plutella xylostella. Several extracts were also assessed against two termite species, Nasutitemes exitiosus and Coptotermes frenchi. Bioassay parameters assessed included acute mortality, antifeedant activity, repellency and effects on growth and development. Only five extracts of extracts showed high bioactivity. One (CP020177) showed high acute dermal toxicity against a number of arthropods tested (LD50 and LD95 values in ppm w/v respectively), including TSM (3797, 8108), DBM (1259, 2185), GHT (3196, 10701) and N. exitiosus (813, 1393). These investigations suggest there is potential for the development of novel pesticides from Australian flora.

Pesticide resistance in Brazil & IRAC-BR activities: current situation & perspectives.

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Recognition of pesticide resistance problems in Brazil dates of the late 1960s, but it was not followed by further studies until the 1990s, when the interest was renewed due to reports of insect-pest control failures. The establishment of the Brazilian Insecticide Resistance Action Committee (IRAC-BR) in 1997 was a reflex of this concern and provided new impetus to pesticide resistance studies in the country. The major activities of the IRAC-BR have been centered on institutional, research and educational programs. The institutional program is based on the promotion of the Committee and its activities, while the research program establishes priorities and provides support for projects of interest. At present, the research effort is centered in two long-standing projects, *Tuta absoluta* in tomatoes and *Spodoptera frugiperda* in maize/cotton, and several other shorter-term projects. The educational program provides basic training in Insecticide Resistance to a diverse public. Annually, IRAC-BR offers short-courses, workshops, lectures and round-table discussions in different regions of the country. IRAC-BR also sponsors a National Research Award, in two categories, undergraduate and graduate research projects. This award was created to stimulate creativity, positive criticism and human resource formation in Insecticide Resistance Management in Brazil. These initiatives from IRAC-BR have been well received in Brazil and the fruits from this effort are beginning to ripe.

Anopheline insecticide resistance in agroecosystems with high & low insecticide use in Northern Thailand.

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Thailand has during recent decades experienced a diversification of the agricultural sector leading to more pesticide intensive cropping systems. About 80% of the pesticides used are for agricultural purposes. Insecticide use is particularly common in fruit orchards. Recent research from rural areas in northern Thailand indicates that anopheline density and diversity decrease as the area of fruit orchards increase. The conversion of forest to intensively managed agricultural systems may have an immediate negative effect on the abundance of malaria mosquitoes, but the long-term effect might results in highly resistant mosquito populations, due to the selection pressure of agricultural insecticides. In November 2003, mosquitoes were collected by outdoor human landing catches at three locations in a malaria endemic area in northern Thailand. The three locations were a fruit orchard (high pesticide use), a cornfield (low pesticide use), and a village. There was no significant difference in the density of primary malaria vectors between locations although more mosquitoes were collected in the fruit orchard. Preliminary results from bioassays on insecticide susceptibility show a slight possibility for resistance development in the area. There is a high probability that insecticide resistance is conferred by the heavy use of insecticides in the fruit orchard.

Induction & transmission of Bt-tolerance in the flour moth *Ephestia kuehniella.*

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"The use of *Bacillus thuringiensis* endotoxins to control insect vectors of human diseases and agricultural pests is under threat from the possible evolution of resistance in major pest species. In addition to high levels of resistance, produced by receptor insensitivity, several cases of tolerance to low to medium levels of toxin have been reported in laboratory colonies of lepidopteran species. Since the molecular basis of some of these cases of tolerance to the toxin are not known, we explored alternative mechanisms. Here we present evidence that tolerance to a Bt-formulation in a laboratory colony of the flour moth *Ephestia kuehniella* can be induced by pre-exposure to a low concentration of the Bt-formulation and that the tolerance correlates with an elevated immune response. The data also indicate that both immune induction and Bt-tolerance can be transmitted to offspring by a maternal effect and their magnitudes are determined by more than one gene.
"

Population genetics of kdr-type resistance to pyrethroids in the peach-potato aphid, *Myzus persicae*.

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The peach-potato aphid, *Myzus persicae,* is an important agricultural pest in many parts of the world. It is mainly controlled through insecticide applications and has developed strong resistance to most of the major chemical classes as a result of their widespread use. Resistance to pyrethroids is particularly problematic in *M. persicae* and is primarily the result of a target site resistance mechanism termed knockdown resistance (or k). We have found that this is caused by either one (L1014F) or two (L1014F + M918T) point mutations in the domain II region of the para-type voltage-gated sodium channel (the target for pyrethroids). Aphids that carry the double mutation are highly resistant to even the most potent pyrethroids, surviving doses of deltamethrin in excess of 100ppm. To facilitate rapid monitoring for pyrethroid resistance in *M. persicae*, we have developed an allelic discrimination PCR assay to determine the genotypes of the two mutations in individual adults using fluorescent Taqman® MGB probes. We are using this assay to study the origins and spread of the mutations in a diverse range of *M. persicae* populations.

Modelling distributions & richness of ant species in South Carolina, USA.

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Pitfall collections at 243 sites distributed through four physiographic regions and 29 landscape types defined by SCGAP Analysis program were used to build a GAP layer for ant species. The layer models the predicted distribution and richness of ant species in South Carolina, USA. More than 80 species were collected and modelled. Distributions were modelled using physiographic region as a coarse filter. County lines and SCGAP landscape type were used as fine filters. Spatial analysis was conducted to determine the predicted species richness for each physiographic region and SCGAP landscape type.

Bio-Identifier" – A DNA-based diagnostic tool identifying invasive species at the border.

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Invasions of insect pests have significant consequences for the economy, environment and health in New Zealand. Therefore, the ability to perform rapid and accurate identifications of intercepted organisms is an essential tool. One of the major difficulties in the identification of intercepted material is that it often involves insect immature life stages and strains for which reliable taxonomy based on morphological characters is absent. Therefore, methods that enable accurate and rapid diagnosis of taxa, whether they be well-defined species, or biotypes, are of crucial importance. Developments in DNA technology have resulted in a range of molecular techniques applicable to such applications. Much genetic data also exists in public databases, such as GenBank (NCBI), and can be utilised to develop comprehensive databases across wide-ranging taxonomic groups. We have shown, however, that DNA diagnostic techniques developed without consideration of phylogenetic relationships, or without taking into account the diagnostic value of markers, can give false identifications. Another key feature is the ability to taxonomically assign an unknown specimen even if specific DNA data is not available from that particular organism. These needs have resulted in the development of "Bio-identifier", which is a web-based DNA diagnostic tool integrating poweful bioinformatic tools and validated DNA databases, with links to risk assessment profiles and other taxonomic databases.

Spill over feeding on native Myrtaceae by *Uraba lugens.*

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Uraba lugens, a serious pest of *Eucalyptus* (Myrtaceae) in Australia, is now well established in southwest Auckland. Due to concerns about the potential impact of *U. lugens* on native New Zealand Myrtaceae, the survival and development of *U. lugens* was assessed on 11 native species under controlled conditions. In no-choice tests, larvae developed from 6th instar to pupation on 5 of the 11 species tested; only one species (*Metrosideros umbellata*) supported development from egg to pupation. Two sequential field surveys were performed in southwest Auckland to target early and late instar larvae. Within the survey area, all Australian hosts of *U. lugens* and all native New Zealand Myrtaceae were checked for the pest. Of the five native species found in the survey area (n = 441 trees), only *M. excelsa* was attacked by *U. lugens*. In the first survey 1% of the *M. excelsa* were infested (all < 1m from an infested Australian host) and 6% were infested in the second survey (43% < 1m and 86% < 10m from an infested Australian host). These results suggest that damage to native New Zealand Myrtaceae will be caused by wandering larvae and is likely to be limited to areas adjacent to infested Australian Myrtaceae.

On the road to perdition? Invasives & novel plant-insect interactions.

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Whenever, a novel interaction is initiated, i.e. between a naïve host plant and a foreign phytophagous insect, there is the potential for a death spiral ecological trajectory, i.e. where the insect populations increase explosively but the plant's populations plummet because their capacity to compete, reproduce, and survive is severely debilitated by the foreign insect. Although the possibility of such a trajectory for novel plant/insect interactions is often perceived as common knowledge, interactions of this kind are actually very poorly and disparately documented. We analyze several such interactions in North American wooded landscapes and argue that the potential for severe interactions of this type are most likely when the phytophagous insect has an intimate feeding relationship with its naive host plant. Feeding intimacy is defined by the amount and duration of contact between the insect's mouthparts and/or its body and living plant tissue.

Distribution of an indigenous & an introduced biotype of *Bemisia tabaci* (Gennadius) in Reunion Island using microsatellite markers.

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Since 1997, *Tomato yellow leaf curls virus* (TYLCV) has been responsible of great losses on tomato production in Réunion island. *B. tabaci* infestations were not observed on vegetable crops before the detection of TYLCV in 1997, although *B. tabaci* had been reported several times in the island since 1938. After 1997, two genotypes of *B. tabaci* were differentiated in Reunion. One was identified as biotype B and is supposed to have been recently introduced, whereas the other is distinct from any genotype analysed so far. Since the latter genotype was also found in Madagascar, Mauritius and Seychelles, it was considered native from the South west of the Indian Ocean and was named Ms. The distribution of both biotypes was studied throughout Reunion using a genotyping technique based on microsatellites markers. *B. tabaci* was sampled in 30 different locations typical of the various ecosystems found in Reunion. Wherever possible the whiteflies were collected on crops and weeds to investigate an eventual host specificity of the biotypes. The B biotype was mainly found in vegetable crops and its geographic distribution differ from the Ms distribution. Future equilibrium between the invasive and the indigenous biotypes is discussed regarding to virus epidemiology .

Rhagoletis cingulata Loew (western cherry fruit fly) in the Netherlands (Diptera: Tephritidae)

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Rhagoletis cingulata Loew, is known as a serious pest of cherries in the western United States and Canada. It can infest sweet and sour cherries, *Prunus avium* and *P. cerarsus*, respectively and can cause considerable damage. In 1999, a single specimen of *R. cingulata* was trapped in a Malaise trap in the province of Zeeland. This first Dutch record initially was referred to *R. indifferens*, but later it appeared to be the closely related *R. cingulata*. The following years the species was found at several localities in the coastal dune region, exclusively on *Prunus serotina*. Because the species is listed as a quarantine species for Europe (Council Directive 2000/29/EC, Annex IAI) in 2003 a national survey was carried out both in *Prunus serotina* and in cherry orchards. In total 271 Pherocon AM traps were placed, 181 traps in natural environment and 90 traps in cherry orchards. On the yellow sticky traps 3204 specimens of *R. cingulata* were trapped, 3201 specimens in the natural environment and 3 specimens on the traps in cherry orchards. Based on this survey we conclude that *R. cingulata* is established in The Netherlands.

Distribution potential of three *Anoplophora* species in Europe (Coleoptera: Cerambycidae: Lamiinae): a climex study.

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"Numerous exotic invasive species can be inadvertently transported across international borders into regions where they do not already occur via trade in their host plant or host plant commodities. They can also transported as "hitchhikers" with commodities other than their host. Wood-boring species can be carried in solid wood packaging material such as packing cases, pallets, spacers and dunnage, accompanying traded goods. All three *Anoplophora* species (*Anoplophora glabripennis* (Motschulsky), *Anoplophora chinensis* (Forster) & *Anoplophora malasiaca* (Thompson)) have a Asiatic orgin and closely related to one another. The ranges of the three species have increased as a result of widespread planting of their hostplants. As polyphagous, wood-boring insects they have followed the expanding ranges of a wide variety of trees, including *Acer* spp., *Populus* spp., *Salix* spp., *Malus* spp., *Citrus* spp., *Prunus* spp. and *Ulmus* spp. Within China, *A. glabripennis* causes serious damage to *Populus* plantations, whereas *A . chinensis* is a serious pest of fruit- and ornamental trees. *A. malasiaca* is the most dreaded pest of *Citrus* in Japan. Host plants grow across much of Europe and using the computer program CLIMEX, several regions of Europe were highlighted as areas where climate is most suitable for the pest`s establishment. "

Biodiversity: threats for the present, opportunities for the future

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The explosion of world population from 2.5 billion people in 1950 to more than 6.3 billion now, coupled with rising standards of affluence and expectations for consumption and the widespread use of inappropriate technologies, has changed the condition of the world from a sustainable condition only 30 years ago to an unsustainable one now. In other words, all ecological indicators, taken on a global scale, trend downward. Indeed, it has been estimated that it would take the productivity of two additional planets equivalent to Earth if all humans were to enjoy the standard of living characteristic of Australia and other industrialized countries. This clearly indicates that population, affluence, and technology must be addressed to find global stability. Against such a background, we are struggling to learn more about biodiversity, and believe that the 21st century should be a time when our deepening knowledge of biology gives us the ability to construct a sustainable world in which starvation and poverty will be alleviated. However, even by generous standards, we have catalogued no more than a sixth of the eukaryotic organisms on Earth, and very probably an even smaller proportion of the insects. We understand in any detail only a small proportion of those to which we have given names, and are moving to deal with the rest mostly in ways that would have been more suitable for the 18th century than for the 21st.

Efforts in Australia have led the world in learning about local biodiversity, but are coming under increasing competition for funding as other priorities increasingly dominate Commonwealth and State budgets. The relatively modest investments made in this country and elsewhere have demonstrated huge potential to assist greatly in the attainment of sustainability and social justice throughout the world, and it appears to be sure folly to neglect the opportunities that are available to us now, which are greater than any that we shall have in the future. Continued exploration, the more widespread use of databases, and conservation strategies fit for a rapidly changed and already seriously altered world must be adopted for the sake of future generations.

Insects: a class act through history

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There is perhaps no impending ecological crisis about which the world cares less than the potential extinction of insect species. Even science fiction writers who routinely mine the environmental science literature for film and novel scenarios find the prospect to have little dramatic potential, despite the fact that a considerable body of evidence attests to the vulnerability of insects and other arthropods to population decline. Insect extinction is a reality; at least 72 species are listed as extinct worldwide by the International Union for Conservation of Nature and Natural Resources (IUCN). This tiny number, relative to the number of described species, is reflective more of the lower level of concern for their welfare in general and the greater degree of difficulty in documenting their population declines than it is any extraordinary immunity from extinction on the part of arthropods, in comparison with other organisms. What few comparative studies have been done in fact suggest otherwise. Due to the tremendous trophic diversity of insects, extinctions within other taxa can be accompanied by unrecognized (or at least unheralded) insect extinctions. Although the extinction of many bird species, for example, has been meticulously documented, the status of their arthropod ectoparasites is rarely documented, much less mourned. Making the case for protecting the blood-sucking parasites of a more charismatic vertebrate struggling for survival is perhaps the ultimate challenge in conservation biology.

hy conservation of arthropods, and unappealing invertebrates in general, is such a hard sell derives in large part from a general lack of appreciation of the enormous impact of insect goods and services on human health and welfare throughout history and into the present. The consequences of extinction in select taxa are potentially of tremendous economic importance (and certainly on a par with those of more frequently exploited science fiction scenarios). Enlightening policymakers and the public of the necessity for continued cohabitation of the planet with insects is among the greatest challenges faced by contemporary entomologists and may involve new knowledge, new vocabulary, and new outreach efforts befitting the new century.

Documenting ecosystem services provided by insects, an effort used with increasing frequency to promote conservation efforts in other groups, has rarely been attempted and presents a set of unique challenges. Such efforts are inevitably hampered by the paucity of data on life histories and ecologies, particularly for species that have experienced drastic reductions in population size. The same can be said, perhaps even more forcefully, for arguing from the perspective of biodiversity prospecting. Substances of pharmaceutical value may be difficult to extract or produce in economically viable amounts given the small size of the producers and the difficulties associated with mass rearing.

Moral and aesthetic arguments for conserving insects have rarely met with the same level of success as they have with more conspicuous, majestic, or symbolically powerful species. Outreach efforts in the context of insect conservation should be sensitive to the power of language. The value of an accessible and appealing common name, for example, particularly one that reflects distinctive life history attributes, is inestimable. The best hope, though, for success is for entomologists as a group to make every effort to reach out to the public to cultivate an appreciation for the Class Insecta; there are more than a million stories and they are all ultimately worth telling.

"Honeybee vision, navigation and 'cognition': Basic principles and technological applications"

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Insects, in general, and honeybees, in particular, perform remarkably well at seeing and perceiving the world and navigating effectively in it, despite possessing a brain that weighs less than a milligram and carries fewer than 0.01% as many neurons as ours does. Working together with our colleagues, we have been trying to unravel the secrets of their success.

Although most insects lack stereo vision, they use a number of ingenious strategies for perceiving their world in three dimensions and navigating successfully in it. For example, distances to objects are gauged in terms of the apparent speeds of motion of the objects' images, rather than by using complex stereo mechanisms. Objects are distinguished from backgrounds by sensing the apparent relative motion at the boundary. Narrow gaps are negotiated by balancing the apparent speeds of the images in the two eyes. Flight speed is regulated by holding constant the average image velocity as seen by both eyes. Bees landing on a horizontal surface hold constant the image velocity of the surface as they approach it, thus automatically ensuring that flight speed is close to zero at touchdown. Foraging bees gauge distance flown by integrating optic flow: they possess a visually-driven "odometer" that is robust to variations in wind, body weight, energy expenditure, and the properties of the visual environment.

Integrated pest management in the age of biotechnology: how can we be better off in the long run?

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"A "pest" in "integrated pest management" or IPM subsumes any living organisms that bother people. Here we focus on insect pests in crop systems. First we review briefly the origin and development of the IPM paradigm. Next we present a series of outstanding case histories to illustrate how successes and failures of IPM have been brought about. We then use these case histories to pose a few key questions as to how we can improve IPM. Finally we take a glance towards the future of IPM in the age of biotechnology.
Recent research on honeybee perception and cognition is beginning to reveal that these insects may not be the simple, reflexive creatures that they were once assumed to be. For example, bees can learn rather general features of flowers and landmarks, such as colour, orientation and symmetry, and apply them to distinguish between objects that they have never previously encountered. Bees exhibit "top-down" processing: that is, they are capable of using prior knowledge to detect poorly visible or camouflaged objects. They can navigate through labyrinths by learning path regularities, and by using symbolic signposts. They can learn to form complex associations and to acquire abstract concepts such as "sameness" and "difference". Bees are also capable of associative recall: that is, a familiar scent can trigger recall of an associated colour, or even of a navigational route to a food location. All of these observations suggest that there is no hard dichotomy between invertebrates and vertebrates in the context of perception, learning and 'cognition'; and that brain size is not necessarily a reliable predictor of perceptual capacity. Finally, some of the above principles – especially those that relate vision and navigation – are offering novel, computationally elegant solutions to persistent problems in machine vision and robot navigation. Thus, we have been using some of the insect-based strategies described above to design, implement and test biologically-inspired algorithms for the guidance of autonomous terrestrial and aerial vehicles.

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Key elements of the IPM paradigm include: (1) pests are biotic components of ecosystems capable of inducing stress and reducing crop yield or quality, and management activities must envisage pests within such an ecological perspective and work in harmony with natural control; (2) multiple tactics such as cultural control, plant resistance, biological control or physical control are used in a compatible manner, and most importantly naturally occurring biological control should be promoted to the greatest extent possible and application of broad-spectrum synthetic insecticides be minimized, to maintain pest damage below the economic injury level; (3) creatures dubbed key pests are allowed to survive at low abundance in the ecosystem; and (4) cost/benefit analyses must take into account the interests of and impacts on producers, society and the environment. This IPM paradigm is considered to be the best science available to the solution of pest problems, and no major departures from the basic research and implementation paradigm have occurred in the past 40 years, although significant changes have taken place in technological advances and tactical options available. Nevertheless, ecological research findings and IPM implementation experiences over the years have contributed significantly to providing strong support to the underlying principles and improving the elaboration of details.
Examples are used to illustrate the successful and unsuccessful approaches that have been used to develop and implement pest management programs. The final victory of the vedalia beetle and associated drama demonstrate that (1) an introduced pest can be effectively controlled by the right natural enemy introduced from the pests origin; (2) the beauty of naturally occurring biological control can be immediately devastated by a humanistic approach, and (3) to follow blindly a correct IPM idea, without adequate ecological research, can be unproductive. The history of migratory locust control in China records that habitat manipulation, not chemical control, solved, perhaps for good, the worst pest problem that has devastated the life of Chinese people for thousands of years. Rice IPM in Asia illustrates that (1) even in annual crop systems naturally occurring biological control plays a vital role in suppressing the pests; (2) a participatory approach among farmers, extension service and researchers is needed to effectively implement IPM; (3) sound ecological research is necessary to avoid misuse of seemingly powerful tactics; and (4) forceful legislative measures can be a powerful tool to aid in IPM implementation. Experiences in cotton IPM suggest that practical IPM strategies can be derived only through an experimental approach, and (2) extensive and expensive strategic modelling with computers has not provided much insight of use to practical IPM. The recent successful *Bemisia* IPM in the United States highlights that a 'super-bug' can be brought under sustainable control by IPM, mainly through a right integration of biological and chemical controls. The experiences of brassica IPM in New Zealand, Australia, and China illustrate that (1) classical biological control can form the central element of effective pest management programmes; (2) effective naturally occurring biological control can be promoted by simple IPM strategies; and (3) field comparative experimental studies can offer direct and convincing evidence of the efficacy of biological control. The commercialized successes of augmentation biological control in greenhouses in Europe demonstrate that even in highly artificial crop systems, biological agents can be promoted to bring pests under control and the approach is sustainable. The recent history of deployment of transgenic insecticidal plants indicates that (1) insect resistance to these crops in the field can be effectively delayed by appropriate resistance management strategies, and these plants offer new opportunities for integrating plant resistance with biological control to develop more successful IPM programs.
Philosophically there are two conflicting approaches to IPM, one rooted in naturalism and the other in humanism. The key

elements of the IPM paradigm proclaim it as naturalism. To follow a philosophy of naturalism means that we must work with nature whiling trying to manipulate its features for our benefits. The case histories demonstrate that as we become equipped with more powerful tactics, we need to use them more gently and intelligently to avoid disasters and to gain the best benefits in the longer term. We assert that the IPM paradigm, as it has evolved to date, has proven to be a robust construct for the solution of pest problems. The key weaknesses with our pest control research and practices in recent years have come from a habitual desire to view IPM simply as a substitute for "pest control", with little thought given to the underlying ecological principles and the implications for maximizing the inherent resistance to insect pests in the ecosystem. Major technological advances in chemistry, biochemistry, behavioural science, neurophysiology, molecular genetics, and genetic engineering can be expected to produce a new array of products and materials that are less toxic and hazardous to humans and the environment than conventional pesticides in the years to come. Deployment of transgenic insecticidal plants and other new technologies guided by the IPM paradigm are likely to enable us to develop sustainable pest management systems that will maximize economic, social and environmental benefits for society in the long run.
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Genetically modified crops and food security: an African perspective

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In Africa 70% of the population depends on agriculture as the sole source of income. However, Africa's crop production is the lowest in the world and 25% of grain is imported. It is estimated that sub-Saharan Africa will have a shortfall of nearly 90 million tons of cereal by the year 2025. Biotechnology and the use of genetically modified (GM) crops are some of the tools that could improve efficiency, quality and productivity. At a UNIDO meeting in Nairobi last year, agricultural experts from some 13 African countries prioritised biotechnology-derived crops as follows: insect resistant (Bt) African maize varieties, resistance to African plant viruses, resistance to parasitic weeds, decreased mycotoxin levels in maize and drought tolerant crops.

Maize streak virus is endemic in Africa, causing huge economic losses to both commercial and small scale farmers. Colleagues and I have developed maize that produce a severely mutated form of the replication associated protein, resulting in highly resistant plants. These are being bred into commercial varieties of maize. A consortium of African and American scientists are developing cassava resistant to the *African cassava mosaic virus*.

A major parasitic weed is Striga, or witchweed. It attacks the roots of poorly growing crops and intertwines its roots with them. Field trials have been carried out on herbicide resistant maize with very promising results.

South Africa recently began planting Bt white maize for human consumption and we are finding a decrease in the levels of post-harvest fungal infection with a concomitant decrease in mycotoxin content. Bt cotton, although not a food crop, is providing a much-needed increase in revenue to small scale farmers.

The source of our genes for the development of drought tolerant maize is a South African indigenous "resurrection plant" which can loose 95% water content and, upon watering, "resurrect" in three days. Our first transgenic plants are showing resistance to water loss, heat and salt.

While GM crops are not a panacea for feeding Africa, we cannot afford to turn out backs on any technology that could improve food productivity. Indeed, 30% of the global transgenic crop area of 67.7 million hectares in 2003, equivalent to over 20 million hectares, was grown in developing countries. The absolute growth in GM crop area between 2002 and 2003 was almost the same in developing countries (4.4 million) and industrial countries (4.6 million hectares), with the percentage growth more than twice as high (28%) in developing countries compared with industrial countries (1.1%). It is clear that farmers, who in Africa are so often also consumers, know a good thing when they see it.

Insect symbionts – life with multiple genomes

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Vertically transmitted microbial symbionts of insects have been painstakingly observed and recorded for well over a century. Not until quite recently though has the true extent of the ubiquity of these associations been fully realized. Similarly a mechanistic understanding of how these agents interact with insects and their importance to the biology of their hosts has only very recently begun to be appreciated. Two broad classes of symbionts are present in insects. The classical nutritional mutualists that provision host insects with required metabolites (e.g. *Buchnera* of aphids and *Wigglesworthia* of Tsetse) and the reproductive parasites that manipulate the reproductive biology of their hosts to maximize their own transmission often at the expense of host fitness (e.g. *Wolbachia* and *Cardinium* that infect a broad range of different insect species).

The recent sequencing of a number of diverse insect symbiont genomes is beginning to peel away the mystery that surrounds these interactions. The completion of full genome sequences of the nutritional symbionts of aphids, tsetse flies and carpenter ants together with the sequencing of the widespread reproductive parasite genome, *Wolbachia pipientis,* is providing insight into the mechanisms that allow these microorganisms to infect and interact with hosts. We now understand the types of molecules that nutritional symbionts provide to their hosts, how they are delivered and the selective pressures that have shaped these associations. For the first time we can start to assess the potential mechanisms by which reproductive parasites manipulate insects and generate host phenotypes like cytoplasmic incompatibility, parthenogenesis, feminization and male-killing.

Together with an understanding of the basic biology of host-symbiont interactions has come the potential to utilize symbiont systems for applied outcomes. Paratransgenesis has emerged as an exciting possibility for modulating the ability of insects to transmit pathogens to humans, plants and livestock. For example, the engineering of bacterial symbionts of *Rhodnius proxilus* to prevent this insect from being capable of transmitting trypanosomes to humans is well developed as a potential future disease control strategy. In addition *Wolbachia* is being developed as an agent that might similarly be used to either suppress insect populations or modify their disease transmission potential. In this case its ability to rapidly invade host populations and skew insect population age structure are features that might facilitate its future use in applied entomology.

Genetic basis for variability of Cry1Ac expression among commercial transgenic *Bacillus thuringiensis* (Bt) cotton cultivars in the United States

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The amount of Cry1Ac in transgenic *Bacillus thuringiensis* (Bt) or Bollgard® cotton varies among commercial cultivars. These expression differences have been correlated to survival levels in Lepidoptera, indicating that all Bollgard® cultivars do not provide the same level of control. The objective of this study was to determine if differences in expression among commercial cultivars of Bollgard® cotton were under simple genetic control. If so, these findings could influence the way breeders select cultivars by selecting for efficacy in addition to agronomic traits. Crosses were made in the greenhouse with a high expressing and a low expressing cultivar. The parents, F_1, and F_2 generations were planted in the field. The amount of Cry1Ac was quantified using a commercial ELISA kit. Highly significant variances within the two F_2 breeding populations were due to genetic segregation for Cry1Ac expression. Using the modified Castle-Wright formula, estimation of the number of contributing genes in both breeding populations was small. These data show that genetic background has a major effect on Cry1Ac expression. Because backcrossing is the primary method used by commercial cotton breeders, the selection and use of donor and/or recurrent parents that will result in a high level of Cry1Ac expression is crucial.

Wild and cultivated sesame (*Sesamum* spp.) resistance to *Antigastra catalaunalis* (Dup.) (Lepidoptera: Pyralidae)

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Sesamum indicum L. and wild relatives (*S. alatum* Thonn., *S. laciniatum* Klein., *S. mulayanum* Nair, *S. malabaricum* Burm., *S. radiatum* Schumach & Thonn) were evaluated in the laboratory (27 ± 0.5 ^{0}C, 75 % RH) for resistance to *Antigastra catalaunalis* (Dup.). On sesame (*S. indicum*) the larval period and the larval to adult periods averaged 12.6 and 17.6 days, respectively, compared to larval periods of 9.33 - 9.75 days and larval to adult periods of 15.5 - 16.0 days on wild species. Pupal periods were similar on all species. Larval survival ranged from 10.25% on S. *malabaricum* to 35% on *S. alatum*. S. *malabaricum* did not support larval growth (no adult emergence), whereas adult emergence ranged from 05.55 - 11.76% on other wild species. Adult emergence was 80% on cultivated sesame. Growth indices and survival index were highest for larvae reared on *S. indicum*. Fourth instar larvae inflicted little feeding damage (0.0 to 05.0 %) to seeds of the wild species in the laboratory. No damage was observed on wild species in the field. This research has identified a source of *A. catalaunalis* resistance in wild *Sesamum* spp

Decrease in the Colorado potato beetle densities on manure-amended soils: another proof of the mineral balance hypothesis?

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The mineral balance hypothesis suggests that the organic matter and microbial activity associated with organically managed soils affords a buffering capability to maintain nutrient balance in plants. An optimal nutrient balance, in turn, results in both good plant growth and resistance to herbivory. We determined effects of soil amendment practices on the Colorado potato beetle populations in potato fields and investigated their interactions with crop rotation and pest management approach. Beetle densities were almost universally lower in the plots receiving manure as soil amendment, while crop rotation and pest management approach had little or no effect. Unlike beetle abundance, plant height and canopy cover were comparable between the plots receiving manure and synthetic fertilizer. Furthermore, tuber yields were higher at the manure-amended plots. In direct accordance with the mineral balance hypothesis, there was a amatic dissimilarity in the mineral composition of potato leaves collected from manure- and synthetic fertilizer-amended plots. Overall, we detected differences in concentrations of nitrogen, calcium, magnesium, phosphorus, aluminum, boron, copper, iron, manganese, and zinc. Mineral content of potato foliage was responsible for 40-57% of the variation in the Colorado potato beetle populations observed among the experimental plots.

Major insect pest shifts following the adoption of bollgard cotton in North Carolina, 1996 to 2003.

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In an 8-year, large-scale evaluation of pest shifts resulting from the adoption of Bt (Bollgard) cotton by North Carolina's cotton producers, 623 pairs of Bollgard and conventional cotton fields were assessed from 1996 through 2003 for mid- to late-season boll damage. Bollworm, *Helicoverpa zea* (Boddie) damage to bolls averaged approximately 3-fold higher in conventionally protected cotton (4.7% vs. 1.7%), while stink bug, mostly *Acrosternum hilare* (Say), damage to bolls averaged more than 3-fold higher in Bollgard cotton (3.0% vs. 0.9%). European corn borer, (*Ostrinia numbilalis* (Hübner), and fall armyworm, (*Spodoptera frugiperda* (J.E. Smith), damage to bolls was very light from 1996 to 2003. Overall boll damage during this period favoured Bollgard cotton by 1.3% (4.9% vs. 6.2%). Bollgard cotton was treated an average of 2 times less per year (2.65 vs. 0.85 applications) than conventional cotton. Considering the average technology fee for Bollgard cotton seed, late-season insect control costs, the penalty of boll damage, and proposed relative scouting costs: the overall economic returns of Bollgard cotton (for insect-related costs only), averaged over the 1996 to 2003 commercial availability period, were approximately $15.00 (US)/hectare greater than that of conventionally protected cotton.

Management of forests for the future - site selection and environmental change

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1. The major part of the European forests originate from a strong modification of the native forest ecosystems, mainly based on the introduction of conifer species from outside their natural range. 2. Most of the literature on forest insect pests concerns tree species considered as economically importantconomic importance can be related either to the cost of the investment or to the expected benefits. 3. The forests subjected to insect attacks may vary according to the climate change, as it may affect the distribution of the insect pests to a greater extent than that of the host trees. 4. The environmental change affects indirectly the tree-insect relationships because of the new functions of the forests (e.g. CO2 sink, renewable energy sources, touristic use) which may results in new management options. 5. Little is known about stability mechanisms of low- or no-disturbed patches of European forests, which could be potentially used to assess the resiliance of the pristine habitats to the environmental change. 6. Extreme events, such as the high temperature in the summer of 2003, may allow 'a posteriori' test of hypotheses on the consequences of the climate change on the tree-insect relationships. 7. A few management options (e.g. use of trees within the species range, mixed versus pure stands, improvement of the understory) have been proposed to counteract the effects of the environmental change, but there is limited experimental evidence supporting each of the suggested options.

Use of mineral oils in tropical and subtropical citrus IPM in Asia: potential and impediments

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Extensive studies since 1993 have shown significant potential for using biorational horticultural and agricultural mineral oils in citrus IPM programs in Asia. We summarise results that show that these products can be used to control a wide range of pests and diseases simultaneously in programs based on application of multiple evenly and adequately (generally low volume) applied low-concentration (£ 0.5% v/v) sprays. We show that these programs are cost effective and that they can be used to significantly reduce currently excessive use of broad-spectrum synthetic pesticides. We discuss impediments to widespread adoption of this technology and means of overcoming them. These include poor farmer education, 'quick kill' and 'the only good insect is a dead insect' paradigms, chemical companies and their distributors, marketing of relatively cheap, substandard highly phytotoxic petroleum-derived sprays oils, government policies, regulations and their enforcement, and incompatibilities stemming from inappropriate overuse and mixing of pesticides, fertilisers and emulsifiers.

Mapping stem borer and armyworm resistance and putative resistance factors in tropical maize.

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Maize production in tropical ecologies can be severely reduced by stem borers s (*Diatraea spp.*) and fall armyworm, *Spodoptera frugiperda* (J.E. Smith). The International Maize and Wheat Improvement Center (CIMMYT) as developed tropical sources of resistance to these pests for use in the development of molecular maps to identify quantitative trait loci (QTL) for the observed resistance. Using two molecular maps [CML131 (S) x CML67 (R) and Ki3(S) x CML139(R)], QTL were identified using composite interval mapping. Five major QTL in bin 1.06, 7.03, 8.02/6, and 9.03 explained 41% of the phenotypic variance (σ^2_p) for leaf feeding damage. Three QTL for leaf toughness, a trait considered important in resistance, were located in bin 1.06/11 and 8.06 and explained 37% of σ^2_p. Chlorophyll QTL did not overlap with insect resistance factors, with the major QTL being located in 4.08 ($\sigma^2_{p=}43\%$). QTL for cell wall bound phenolic acids (*trans*-ferulic acid, 8-5′ diferulic, and 8-5′ diferulic acid) clustered with resistance in bin 1.11. This observation is consistent with a broad-based, structural resistance mechanism for tropical maize resistant to lepidopteran field pests. Based on these studies, candidate genes have been identified and will be discussed.

Understanding aphid damage and development in canola crops

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Canola crops grown for oilseed in Western Australia are regularly infested by the aphids *Brevicoryne brassicae* (L.), cabbage aphid, *Lipaphis erysimi* (Kaltenbach), turnip aphid, and *Myzus persicae* (Sulzer), green peach aphid. Research into whether economic damage results from these infestations has been under way for a number of years. Early work showed that high levels of aphids in canola did cause temporary damage, but these infestations did not translate into losses in oilseed production. This was explained by the ability of canola plants to compensate for insect damage. Canola's ability to compensate for aphid damage is influenced by rainfall and to a lesser extent, plant cultivar. In addition, the level of plant drought stress influences the rate of aphid population development on canola. Aphids develop at faster rates on susceptible cultivars of canola when plants are undergoing drought stress than on unstressed plants, and will eventually cause significant and irreparable losses in oilseed production. Understanding the influence of these factors on oilseed production will aid in the decision making process for aphid control measures. The details of this research are discussed.

Recent advances in the management of *Rhopalosiphum padi*, a vector of barley yellow dwarf luteovirus

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The bird cherry-oat aphid, *Rhopalosiphum padi* (L.) (Homoptera: Aphididae) is one of the most economically important aphids on wheat worldwide. *R. padi* damages the host by direct feeding and by transmitting *Barley yellow dwarf virus* (BYDV). This luteovirus causes a serious disease of cereals worldwide. Management of *R. padi* is achieved via multiple practices including chemical, cultural and biological control, but relies heavily on insecticides. Seed-applied insecticides can reduce aphid densities and BYDV incidence. Planting date modifications may be used to avoid aphid flights and reduce BYDV incidence. However, such modifications are not always a practical management option. Natural enemies, including parasitoids and predators provide aphid control and may influence virus spread. Models have shown that the effect of vector preference for virus-infected plants on virus spread is influenced by the frequency of diseased plants. We have recently demonstrated that *R. padi* behavior is influenced by BYDV infection of wheat plants and that this is mediated by plant volatiles. Knowledge of the factors that influence *R. padi* responses to BYDV-infected plants would be useful for understanding vector behavior and movement, and virus spread. The potential implications of these findings for the development of management options will be discussed.

Efficacy of a pyrethroid and a systemic seed-treated insecticide to manage a pest and a pathogen

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Bean pod mottle virus causes qualitative and quantitative damage to soybeans and is transmitted most efficiently by the bean leaf beetle, *Ceratoma trifurcata* Forster. During the summers of 2002, 2003 and 2004, potential strategies for managing bean leaf beetles and *bean pod mottle virus* were evaluated at Iowa State University research farms. The current management strategy in Iowa recommends one early and one mid-season lambda-cyhalothrin application. Our study determined the efficacy of applying a systemic, seed-treated insecticide (thiamethoxam), either alone or in combination with foliar applications of lambda-cyhalothrin. Six (2002) and seven (2003 and 2004) management strategies were evaluated. In 2002, the following treatments were used: thiamethoxam seed treatment, lambda-cyhalothrin foliar application in late May, lambda-cyhalothrin foliar application in early July, thiamethoxam seed treatment with a lambda-cyhalothrin foliar application in early July, lambda-cyhalothrin foliar application in late May and early July, and an untreated control. In 2003 and 2004, one treatment was added — thiamethoxam with a lambda-cyhalothrin foliar application 10 days later. The effectiveness of these strategies was measured by comparisons of beetle abundance, enzyme-linked immunosorbant assayed soybean sap, seed weight and quality.

Invasive ant species in continuous and fragmented areas in the Atlantic forest

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We investigated the role if invasive ant species along the Atlantic forest, one of the world biodiversity hot spots, taking *Wasmannia* spp. as a model organism representing the invasive species. We based our study on fifty 1m^2 litter samples, submitted to Winkler extractors for 48 hours, collected in each of 26 regularly spaced localities along some 20^o degrees of latitude, representing two main classes of altitudes (sea level and 800-900m). We concentrate collecting in the evergreen dense forest in the wet season. As localities in the southern half on the forest are continuously covered by forest, while in the northern half the forest is fragmented, we investigated whether the fragmented half bears significantly more invasive ants than in the continuous forest. For the northern half,we investigated if there is a significant correlation between fragment size and the invasive ants present in the samples. We studied also the effect of the presence of invasive species in each sample regarding the total number of ant species per sample.

A novel approach for rapid screening of grapevine rootstocks for resistance to grape phylloxera, *Daktulosphaira vitifoliae* Fitch (Homoptera: Phylloxeridae).

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Quarantine and the use of resistant rootstocks are currently the only long-term options for managing the viticultural insect pest, grape phylloxera, *Daktulosphaira vitifoliae* Fitch (Homoptera: Phylloxeridae). Phylloxera management in Australia is unique because most commercial vineyards are planted with ungrafted European grapevines and the focus of management is on quarantine to restrict spread of the insect. Most viticultural regions are therefore susceptible to infestations and effective rootstock recommendations need to be developed. Conventional approaches to screening for phylloxera resistance include laboratory and field-based studies. A new protocol was developed to screen grapevine rootstocks for phylloxera resistance under glasshouse conditions. Grapevines grafted to three rootstocks of American parentage used in south-eastern Australia were screened for resistance to the two most common phylloxera genetic strains, G1 and G4. The rootstocks were significantly more resistant to both strains of phylloxera than the ungrafted vines. Population development was significantly reduced on all rootstocks. Furthermore, populations of the G4 strain were significantly greater than the G1 strain on ungrafted roots, indicating different degrees of virulence between the strains. This screening technique is considered to be more reliable than excised-root bioassay systems, which exclude a whole plant response, and field-based studies where external variables can influence results.

Towards appropriate *Bt*-Corn IRM strategies for the Asian corn borer, *Ostrinia furnacalis* (Guenee) in major corn growing areas in the Philippines: field surveys of alternate host plants and biological studies

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The institution of refugia as conventionally defined for temperate areas accompanies the introduction of Bt-corn into the Philippines. The refugia concept has underlying assumptions based on population dynamics, which, in turn, rely also on plant-insect relationships, including alternate hosts. Our survey showed tomato, pepper, sitao, okra, cotton and sugarcane as alternate crop hosts and aguingay, napier, tigbi, paragrass and wild sorghum as weed hosts. The abundance of these plants in and out of the cornfield was noted. ACB completed its development on aguingay, napier and tigbi using fresh stalks under laboratory conditions. Several laboratory experiments determined larval and ovipositional preference. Larvae preferred to feed and adults preferred to oviposit on corn, napier, aguingay and cotton. On the other hand, ACB completed it growth and development on whole plants of all selected alternate hosts except tomato and pepper, which appear to be transient hosts only. Further investigation of other hosts and their relationships with ACB in different cropping systems is recommended. Under tropical conditions, the natural great plant diversity may necessitate changes in, if not a total re-thinking of the refugia concept as an insect resistance management strategy.

Evaluation of plant resistance, chemical, and cultural control to manage populations of the sunflower stem weevil and impact on natural enemies

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The sunflower stem weevil, *Cylinocopturus adspersus* (LeConte) (Coleoptera: Curculionidae), is a pest of cultivated sunflower. Since the 1990s, damage has been reported and populations have been increasing in the sunflower production regions of Colorado and Kansas. Adult weevils emerge from overwintered stalks and females lay their eggs at the base of sunflower stalks. Larvae feed apically in the stems and then descend to the lower portion of the stalk and excavate overwintering chambers into the stem cortex. If the larval population in a plant is high, the stem, weakened by tunneling or overwintering chambers, will break causing a loss of the entire plant. The purpose of this 3-year project was to investigate tactics including a combination of planting date and foliar and seed treatment insecticide applications to lower weevil densities in the stalks and thus reduce losses caused by lodging. Trials included chemical application based on both plant stage and degree-day models. A second goal was to investigate genetic resistance as a potentially valuable resource and screen available sunflower hybrids, accessions, and lines for tolerance to lodging or low weevil densities in the stalks. The impact of these methods on weevil parasitoids which attack the larvae also was studied.

Examining the origins of insect pests on rice: effects of rice domestication on tritrophic interactions

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Many insect pests on rice only feed on the genus *Oryza*, suggesting that prior to domestication, these insects fed on the wild progenitors or relatives of cultivated rice. While rice domestication has increased its suitability to cultivated conditions, other physical and chemical plant traits have also been altered that may affect interactions between rice, insect pests, and their natural enemies. Understanding how domestication has altered tritrophic interactions may help determine why some rice pests are so difficult to control and which plant characteristics can help control pest populations. We investigated the role of rice domestication on tritrophic interactions by comparing the abundance of key rice pests (stemborers, planthoppers, leafhoppers) and their natural enemies on *O. sativa* and *O. rufipogon*, widely considered the wild progenitor of cultivated rice. Plots were sampled for insects on a bimonthly basis using vacuum sampling and destructive sampling. In addition, we measured physical traits such as plant allocation, tillering, stem diameter, and timing of plant growth. We examined whether the patterns of pest and natural enemy abundance were related to physical plant changes associated with rice domestication.

Impacts of Bt transgenic rice on arthropods in rice ecosystem

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Impacts of Bt transgenic rice on community structure of arthropods in rice ecosystem was investigated in isolated rice paddy and the results showed that the abundance and species richness in Bt rice (KMD1) were little higher than those in the control (XS11). Three varieties of Bt rice (KMD1, KMD2 and T7) with the same Bt gene (*Cry*1Ab) and Ubiquitin, but two parents (XS11 and T6) were selected to study the impacts of Bt rice on population parameters and the relevant mechanism in laboratory. The results indicated that the impacts of Bt rice on population parameters of insects varied with insect species and rice variety. The comparisons of free amino acids and plant volatiles among KMD1, KMD2 and XS11 showed that some chemical components in the three varieties were significantly different and the differences might consist with the impacts of Bt rice on biological parameters on the arthropods. Finally, the interactions among main components related to ecological risks in rice ecosystem and the procedures for ecological risk evaluation were discussed.

An overview of citrus IPM in Florida

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Florida citrus production includes ca. 323,000 hectares and represents 70+% of the total United States production. Over 90% of the oranges and 40% of grapefruit are grown for processing and the remainder (30 million or more boxes) goes for fresh market. Many citrus species and varieties are grown and production practices differ depending upon their market destination, susceptibility to fungal pathogens, and insect or mite pests. Florida's climate is best characterized by having both moderate temperatures and rainfall and infrequent freezes. Because of environmental conditions, fungal pathogens are among the most important problems facing Florida citrus growers today. Fungicides are regularly applied and their use patterns, in some cases, can exacerbate pest mite pressure in many citrus varieties. In general, the fungal pathogen, *Mycospharella citri* Whiteside and two species of rust mites (e.g. *Phyllocoptruta oleivora* (Ashmead) and *Aculops pelekassi* (Keifer)) are considered key pests on foliage and fruit, respectively. Current management programs for both fresh and process fruit are presented along with their arthropod pests. Within recent years there has been increasing usage of HMO (horticultural mineral oil) programs to reduce production costs. Added benefit of HMO programs provides reduced pest mite pressure on Florida citrus.

Consequences of grass-endophyte associations on insects

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Fungal endophytes (*Epicholë* and asexual *Neotyphodium* forms) in C_3 grasses have been well studied for 20 years, with many studies focusing on their role in determining the outcome of grass – insect encounters. Research shows that both insect deterrence and toxicity can result from the production of specific alkaloids by grass – endophyte associations, while other research demonstrates that many grass – endophyte interactions have neutral effects on insects. In this symposium talk, we discuss the diverse effects of grass – endophyte associations on insect behavior and survival and our ability to use these associations in agricultural environments. New field research in New Zealand suggests that *Neotyphodium* in meadow fescue deters feeding by root-feeding grass grub (*Costelytra zealandica*). Earlier New Zealand research showed that a chemical (peramine) in endophyte-infected perennial ryegrass acts as a powerful feeding deterrent to adult Argentine stem weevil (*Listronotis bonariensis*). New U.S. research illustrates the toxic and neural effects of endophyte infection in grasses on insects by demonstrating the influence that wild barley species/genotype and endophyte strain can have on the expression of cereal aphid and Hessian fly (*Mayetiola destructor*) resistance.

Susceptibility of blueberry cultivars to the borer *Orthorhinus cylinirostris* (Fabricius)

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Subtropical cultivars of blueberries, *Vaccinia* species are cultivated on a 350 hectare site near Corindi, north of Coffs Harbour in New South Wales. Blueberry bushes in this plantation are infested by the larvae of the elephant weevil, *Orthorhinus cylinirostris* (Fabricius), an Australian native weevil. The larvae tunnel in the crown of the bushes, shortening the productive life of the plants from 20 years to less than 7 in the most susceptible cultivars. There are two groups of cultivars, Southern Highbush hybrids and Rabbiteye, with the former the more susceptible. However, both groups show considerable variation and some Rabbiteye cultivars are relatively susceptible. There are two components to the cultivar susceptibility – firstly the level of larval activity and secondly the response of the plant. The most susceptible cultivars are in the Southern Highbush group and the plants show considerable loss of vigour after infestation by a few larvae, the plants dying within 5 years. Susceptible Rabbiteye cultivars may have been infested by up to 20 larvae, but survive for 10-13 years. The tolerant Rabbiteye cultivars can survive for over 15 years and have only been infested by a few larvae. Cultivar selection is discussed in relation to elephant weevil management.

Mass trapping for control of rice and vegetable pests in South Asia

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Farmers in Bangladesh apply insecticides daily on brinjal, at a cost of 1,000 $US per ha, in an attempt to control the brinjal fruit and shoot borer, *Leucinodes orbonalis* but only apply, on average, one application of insecticide on rice to control yellow stem borer, *Scirpophaga incertulas* per season. This reflects both the relative economic importance of the insect pests and the marketable value of the crops. Nevertheless, it can be argued that *L. orbonalis* has only become an economically important pest because of the intensive use of insecticides. In order to prevent the build-up of resistance to insecticides in rice and to overcome the pesticide-abuse problems associated with brinjal production IPM strategies based on mass trapping with sex pheromones has been developed for control of both pest species and are poised for commercial development in India and Bangladesh. The paper will briefly describe the work undertaken to develop the technology and discuss the prospects for implication at the farmer level.

Insects on native and introduced plants: patterns and potential risks

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When a plant species is introduced into a new environment it is colonised by insects. In the Nordic countries, eighty species of forest insects have thus far been recorded on the lodgepole pine (*Pinus contorta* var. latifolia), a species introduced from North America. The list of species includes insects with the native coniferous tree species Scots pine (*P. sylvestris*) and Norway spruce (*Picea abies*) as their main hosts. The most striking pattern is that herbivorous (needle-, bud-, flower- and cone-feeding) insects come from Scots pine, whereas the saproxylic (feeding on the phloem and xylem of dying or dead trees) come from Norway spruce. Only a few of the insect species considered as pests have caused considerable damage in lodgepole pine stands in the Nordic countries. How the introduction of a plant species affects insect colonisation, insect species diversity (biodiversity), the composition of insect communities, and insect population dynamics (e.g. the risk for outbreaks) will be discussed. In addition, a similar study of insects on an introduced willow species (*Salix viminalis*) and on a native willow (*S. cinerea*) will be presented.

Trap crops for managing *Helicoverpa armigera* in Australian cotton

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Trap cropping is an established technology within Australian cotton IPM. Farmers using trap crops typically deploy approximately 1% of their cotton area to trap crops with the specific aim of diverting *Helicoverpa armigera* and *H.punctigera* eggs away from their cotton crops. The most commonly used trap crops are spring flowering chickpea (*Cicer arietinum*), summer flowering pigeonpea (*Cajanus cajan*) and late summer sorghum (*Sorghum vulgare*). Trap crops are most effective early in the growing season prior to the cotton flowering, and late in the season when cotton is senescing. During these periods the availability of alternative cultivated and wild hosts is often limited, creating a bottleneck for *Helicoverpa* populations. Trap crops are not harvested, and are usually not sprayed with insecticides. Once a significant population of *Helicoverpa* is present within a trap crop, and before adult moths begin to emerge, trap crops are mechanically slashed and the soil cultivated to kill the larvae and pupae within and beneath the crop. Studies on the optimal layout of trap crops consistently demonstrate that blocks attract higher egg densities per unit area than strips or patches. *Helicoverpa* egg densities in flowering cotton did not appear to change with increasing proximity to trap crops. The minimum effective area that should be sown to trap crops to achieve spatial and temporal suppression of *H.armigera* populations is unknown.

Biological control in mix and match forests

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Biological control of forest pests occurs in forests that may be natural, modified, or cropped plantations, employing native or exotic tree species. Pest of concern may be native or invasive. These variables generate a set of eight cases that define the variety of circumstances in which biological control of forest insects may be attempted. Biological control options include natural enemy release and establishment (as either classical biological control or new association biological control), habitat enhancement for natural enemies (conservation biological control), and mass releases of artificially reared natural enemies (augmentation biological control). The applicability and the potential economic benefits, costs, and ecological risks of each of these approaches is largely determined by attributes of the pest (native or invasive), the tree species (native or exotic), and the forest context (native forest, modified forest, tree plantations). The categorization scheme discussed allows for quick understanding of the nature of a pest problem and the general nature and associated risks of potential biological control solutions. Examples of each type of forest pest problem are discussed.

Implications of certification on insect pest management in Australian and South African sustainable forestry

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Worldwide markets are demanding that forest products should come from certified forests, which requires compliance with the certification policies for insect pest management. In 2000 the international Forest Stewardship Council (FSC) declared that: "Forest management shall conserve forest diversity… and maintain the ecological functions and the integrity of the forest." To comply with this, "management systems shall promote the development and adoption of environmentally friendly non-chemical methods of pest management and strive to avoid the use of chemical pesticides." The interim Australian Forestry Standard (AFS) in 2003 declared similar intent; that managers must "maintain the productive capacity of forests" but "reduce reliance on chemicals and favour cost-effective methods that minimise adverse impacts on the environment". The FSC has further prohibited the use of genetically modified organisms as well as persistent and toxic chemicals. When FSC standards are applied to 20 insecticides registered or under permits for use in forestry, only *Bacillus thuringiensis* and spinosad products are acceptable, while diflubenzuron, carbaryl, tebufenozide and imidacloprid may be acceptable. Adherence to certification standards presents many challenges in the short term but also provides incentives for minimising the use of toxic chemicals and developing integrated management of insect pests in a sustainable forest industry environment.

Integrated pest management of sunn pest in west and central Asia

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Sunn pest (*Eurygaster integriceps* Puton) is a destructive insect pest of wheat in West and Central Asia and Eastern Europe. Apart from the direct reduction in yield, the insects also inject chemicals into the grain that greatly reduce the baking quality of the dough. If as little as 2-3% of the grain is affected, the whole grain lot might be rendered unacceptable for baking. About US$40 million is spent each year on pesticides in the Sunn pest-prone areas. The present insecticide-based strategies must be replaced with multi-dimensional integrated pest management (IPM) approaches. In collaboration with its partner national agricultural research systems in West and Central Asia, along with the University of Vermont, USA; CABI Bioscience, UK, ICARDA is developing IPM options for the management of Sunn pest making use of adult and egg parasitoids, entomopathogenic fungi, host plant resistance, mechanical control, and cultural practices, such as adjusting planting date and use of early maturing varieties. The IPM options are extended to farmers through a participatory approach using farmer field schools (FFS) formed around IPM pilot sites established in the different countries in the region. Studies assessing the potential impact of IPM options are an integral part of this research.

Level of infestation of transplanted and direct sown cotton seedlings with insect and mite pests

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Late grown cotton plants are severely attacked by leaf-and boll-worms, aphids, and white flies. Success of growing transplanted cotton would virtually eliminate chemical control of seedling pests. Since very few published work dealt with pests of transplanted cotton, therefore this study was conducted to, mainly, investigate the effect of transplanting on rate of infestation of cotton with seedling pests. Our results revealed that: number of thrips, *Thrips tabaci* Lind; the cotton aphids, *Ahpis gossypii* Glover; and the red spider mites *Tetranychus urticae*(Koch), were significantly lower on cotton seedlings of the nursery beds than that of the permanent field, being 4.6 and 10.7; 1.5 and 5.3; 0.4 and 2.9, in the 2002 season; and 5.7and 13.7; 99.1 and 206.3; 2.7 and 46.2, individual pest/ 6 plants, in the 2003 season , for the nursery beds and permanent field, respectively; and that rate of infestation of direct sown cotton with the cotton leafworm*, Spodoptera littoralis (*Boisduval*), was* significantly higher (number of egg batches increased by 158%) than that of transplanted cotton. Although paper bag seedlings' yield was significantly the highest, labor costs of soil-less seedlings were too much less than paper and plastic bag seedling methods.

Challenges and management requirements for the sustainable use of Bt cotton in IPM systems

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Insects are a challenge for Australian cotton production, with *Helicoverpa* and a diversity of minor pests. Although many components of IPM have been used, synthetic pesticides have been the main control. Pesticide reliance brings environmental liabilities of resistance, off-target ift, non-target impacts and chemical residues. The introduction of Bt cottons in 1996 produced significant reductions in pesticide, and coincided with a resurgence of industry interest in IPM. The combination of Bt cotton and IPM can bring significant gains provided the transgenic technology is carefully managed. Deployment of INGARD^{TM} cotton varieties expressing only Cry 1Ac was accompanied by a comprehensive, pre-emptive resistance management strategy which included: (i) effective refuges (ii) destruction of volunteers and crop residues (iii) planting windows (iv) defined late-season thresholds (v) monitoring of Bt resistance. Pyramided varieties expressing both Cry1Ac and Cry 2Ab (BOLLGARD II) are now commercially available and will provide even greater efficacy and stability for resistance management. While resistance is the greatest risk for Bt cottons their sustained value in IPM systems also requires focus on the management of other pests. Many of these, particularly the sucking pests, are suppressed in conventional cotton by *Helicoverpa* sprays. An expected 80% reduction in pesticide applications in BGII cotton may see such pests resurge. However, beneficial species are also more abundant in Bt crops and may help suppress secondary pests. Maintaining an appropriate balance and retaining the benefits of Bt cottons requires vigilant resistance management, sustained efforts to enhance beneficial species and non-disruptive, short residual pesticides for key sucking pests.

Citrus leaf-eating weevil IPM using soil applied bifenthrin

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Citrus leafeating weevil numbers exceeded 500 per 20 beating sites, and damaged up to 25% of lower leaves in two orchards in Queensland's Central Burnett area. Peak adult emergence from the soil was from mid-October to mid-December. Bifenthrin (10 % in Talstar®) applied to the leaf litter not only suppressed emergence, but within 24 hours of application also killed the beetles already in the tree. Field trials on rate, application timing and water volume showed that 25 ml product in 10L water/tree effectively suppressed emergence throughout the peak period, but at lower rates did so for less than 4 weeks. Rates of 3, 6 & 12.5 ml in 5L of water reduced adult numbers in foliage by 75, 94 & 92%. Recovery was rate dependant - at 4 weeks numbers were 44, 24 & 8%, and at 9 weeks 116, 93 & 36% of controls. Applications at the same rate/m^2 in 2L of water to only one side of the row reduced numbers by 58, 75 & 83%. Thus management decisions can be based on numbers of beetles in the trees at peak emergence, and options are available to destroy them and suppress emergence using a minimally disruptive soil treatment.

Understanding the hessian fly-wheat interaction: an examination of early larval damage to wheat leaves

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The interactions of the Hessian fly with its host plants are characterized by plant defense via R genes and insect adaptation via avr genes. Because of the simple genetics underlying important aspects of this insect-plant interaction, a unique opportunity exists for integrating plant and insect molecular genetics with coevolutionary ecology. Yet, in spite of the relatively well-studied genetic aspects of this interaction, many fundamental questions remain. For example, little is known about how the virulent Hessian fly larva attacks wheat cells and establishes a feeding site. We will report on recent microscopy studies on the early versus middle stages of larval attack. Within the first day of larval attack on leaves of a susceptible wheat seedling, cell walls and cytoplasm are disrupted. Three to eight days after the onset of larval attack, significant ultrastructural damage is apparent at the base of several leaves. Understanding how virulent Hessian flies establish a parasitic relationship with the host plant will provide a strong foundation for understanding how R-gene-defended plants prevent parasitism.

The impact of farm management practices on naturally occurring biological control

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Maximizing the contribution of endemic natural enemies to integrated pest management (IPM) programs requires detailed knowledge of their interactions with the target pest. The impact of the endemic natural enemy complex of the diamondback moth, *Plutella xylostella*, in commercial cabbage crops in southeast Queensland, Australia was evaluated by mechanical exclusion of predators and parasitoids. Pest management practices at experimental sites were scored by analysis of management data and sites categorized as practicing IPM, prophylactic insecticide application or as adopting an intermediate approach. Natural enemy impact was greatest at IPM sites, where insecticide inputs were substantially reduced without decreasing crop yields, and least at sites subject to prophylactic insecticide application. *Plutella xylostella* was attacked by three species of larval parasitoids (*Diadegma semiclausum, Apanteles ippeus* and *Oomyzus sokolowskii*) and one species of pupal parasitoid, *Diaomus collaris*. The most abundant groups of predatory arthropods in pitfall traps were Aranae (Lycosidae) and Coleoptera (Carabidae, Coccinelidae, Staphylinidae). The abundance and diversity of naturally occurring biological control agents was greatest at IPM sites, correlating with the greater *P. xylostella* mortality at these sites. The impact of different pest management regimes on biological control and appropriate strategies to optimize this important natural resource are discussed.

Characterization of *Helicoverpa zea* (Boddie) injury and yield losses in bollgard and bollgard II cotton

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Field experiments were conducted to investigate the impact of bollworms, *Helicoverpa zea* (Boddie), on *Bacillus thuringiensis* (Bollgard® and Bollgard II®) cottons. In the first experiment, individual first instar bollworm larvae were placed into white flowers of conventional, Bollgard, and Bollgard II cottons. Plants were visually examined daily. Bollworm survival and fruiting form injury were recorded. Bollworms injured significantly more squares on conventional cotton than on Bollgard or Bollgard II cotton. The numbers of injured bolls were similar on conventional and Bollgard cotton; however, boll injury was lower on Bollgard II cotton than on conventional or Bollgard cotton. In the second experiment, different levels of white flower infestation by bollworms were established for four durations of infestation. The levels of infestation included 0, 50, and 100 percent of white flowers in 2002. In 2003, the experiment was expanded to also include 10 and 25 percent of white flowers. Bollworms delayed maturity and reduced yields of Bollgard cotton when greater than 10 percent of white flowers were infested for more than one week. Bollworms did not impact maturity or yields of Bollgard II cottons. These data will be important for defining economic injury levels for bollworms on Bollgard and Bollgard II cottons.

Effects of the ecological management of *Acacia mearnsii* plantation residues on soil invertebrate pests in the southern hemisphere

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Limited land for the expansion of wattle plantations in South Africa requires a shift to intensive silviculture of seedlings during regeneration, whereby their survival ensures increased productivity. Indigenous soil invertebrate pests cause noticeable seedling mortality but the effect of different plantation residue management practices on their incidence is unknown. Trials were planted to test different residue management regimes [winowed-burnt-weeded, winowed-burnt-ripped, fallow (mowed, manual weed), winowed-burnt-closer spacing, winowed-'broadcast'-herbicide]. During monthly surveys, damaged seedlings were uprooted to determine the cause of death. Pests included whitegrubs and cutworms that had a higher status than the millipedes, nematodes, grasshoppers, false wireworms, termites and crickets. Cutworm damage was higher in the winowed-burnt-ripped and fallow sites. There was a significantly greater pest infestation on sites where the residue was winowed-burnt-weeded or 'broadcast' (20.34%) than in the winowed-burnt-ripped or fallow (2.36%) treatments. The addition of a rip treatment to the winow and burn regime significantly reduced the pest infestation, especially whitegrubs. Seedlings planted at a closer spacing in winowed and burnt sites also had a lower pest incidence (7.79%). An important management implication is that winow and burn is a standard practice, yet the sustainable silviculture of wattle warrants a move to the broadcasting of plantation residue.

Controlling the lantana bug, *Aconophora compressa* **on fiddlewood trees in Queensland.**

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Aconophora compressa, a sap sucking bug native to Mexico, was released in Queensland and NSW in 1995/96 as a biological control agent for lantana, *Lantana camara*. Within three years of its release the insect was found to have established on lantana, but was also located on a non-target species, fiddlewood, *Citharexylum spinosum*. Fiddlewood is native to Florida and the Caribbean and was widely planted in Queensland in the 1970's and 80's. *Aconophora compressa* numbers were initially low but by 2002-2003 *Aconophora* populations had greatly increased and were causing extensive damage throughout south-east Queensland. The insect continues to spread. Trials of insecticides and application methods have commenced to provide chemical control options to the general public and the arboriculture and horticulture industries. Preliminary results from glasshouse and field trials indicate that imidocloprid provides effective and extended control of this pest. Novel application methods and improved formulations are under consideration to enhance the usability of these chemical treatments for a wider range of tree pests in residential and landscape situations.

Changing relationships among biodiversity, management, and biosecurity in managed and unmanaged forests

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A mounting threat to both natural and managed forests of the world are exotic invasive insects. Several exotic wood-boring insects have colonized North America in recent years, and the rate of introduction will increase with the expansion of international trade. Moreover, the implementation of more stringent shipping rules are unlikely to stem the flow of exotic arthropods into new regions because of the inability of quarantine agencies to keep pace with the volume of transported materials and difficulties in enforcing compliance. Exotic species can not only have severe economic impacts by degrading and killing lumber, crop, and ornamental trees, but also will indirectly influence arthropod biodiversity in natural and managed forests. Exotic wood borers in particular may reduce the abundance and diversity of endemic wood borers, and their natural enemies, by eliminating host plants and by competing for resources. For example, the expected decimation of ash populations by emerald ash borer in North America inevitably will have significant repercussions in communities of wood borers that are ash specialists. The challenge for entomologists now becomes identifying threatening exotic invasive species, developing programs for preventing their introduction and managing established species to minimize their deleterious impact, all in accordance with the activities of national security agencies.

Changes in the driving forces behind pest control strategies in export fruit industries such as the southern African citrus industry

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Tree fruits are an important component of international trade in fresh produce and supplying lucrative markets is particularly attractive for farmers in less developed subtropical regions. Production criteria combined with changing demands of sophisticated export markets, represents a unique set of forces that influence the development of pest control strategies in such regions. These driving forces necessitate particularly innovative solutions and consequently the development of pest control in these industries provides valuable direction to the global development of pest control strategies. As an example, southern African is the third largest citrus exporter and 90% of this industry's income is derived from exports to over 60 countries. Whereas pest control developments in this industry used to be driven primarily by IPM principles, this has been replaced by the necessity to find a balance with market demands as diverse as quality assurance and socio-economic responsibility. Additionally, the need to overcome barriers to trade presented by sanitary and phytosanitary regulations affecting international market access, represents the forefront of current and future iving forces behind the development of pest control strategies. Exporting tree fruit industries will again be forced to be at the forefront of developing innovative solutions to these challenges.

Managing a wheat-mite-virus complex: the need to understand the basics

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The wheat curl mite, *Aceria tosichella* Keifer, transmits two serious viruses, wheat streak mosaic virus and high plains virus, to wheat throughout the Great Plains of North America. But, just in the last year reports have also been made of wheat streak being found in both Argentina and Australia. This virus-vector relationship has been known since the 1950's; however, the identification of this mite has long been confused with that of the dry bulb mite, *Aceria tulipæ* Keifer. Recent developments are beginning to give us a clue as to the complexity and significance of this mite-vector relationship. To begin to answer questions concerning mite identification, we have carefully characterized several wheat curl mite populations. Through genetic analyses, three major 'haplotypes' of wheat curl mites have been identified. This variability parallels the ability of these mites to transmit high plains virus and the populations' response to mite-resistance genes in winter wheat. As we move closer to the incorporation of WSMV resistant germplasm into commercial use, understanding these basic mite-virus relationships becomes more important because of the need for the long term stability of resistant genes in the wheat germplasm.

Communication of pest management recommendations – experience from Asian rice farmers

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Rice farmers tend to spray for visible damages caused by leaf-feeders. However such damages seldom reduce yields because of plant compensation. Insecticide sprays especially in the early crop stages instead disrupt normal food web formation causing secondary pests, such as brown planthoppers. This misconception problem is widespread among Asian rice farmers. Decision-making and ecology knowledge were "distilled" into a simple rule or "heuristic" - "Spraying for leaf feeders in the first 40 days is not necessary". After extensive farmer evaluations, media materials were created to motivate farmers. The campaign, launched in 1994 as a pilot in Vietnam, eventually reached ~2 million farmers. The campaigns were implemented through a multi-stakeholder participatory process to enhance local ownerships. This methodology was adopted in Thailand in 2001where a 22 % reduction in insecticide was achieved and in northern Vietnam in 2002 to achieve a 30% reduction. In the Mekong, "Three Reductions" campaign in 2003 motivates farmers to further reduce pesticides by reducing seed and fertilizer rates. We learnt three lessons; 1. the importance of understanding farmers' decision making; 2. the importance of developing quality local partnerships through a participatory process and 3. that media resources can be utilized creatively to communicate of pest management to farmers.

Rice stink bug problems in Japan: abundance and control

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There are over ten species of rice stink bug (RSB) in Japan. Amongst the most damaging species are *Eysarcoris lewisi* in northern Japan and *Leptocorisa chinensis*, *Cletus punctiger*, *Nezara viridula,* and *Eysarcoris ventralis* in central and western regions. *E. parvus, Trigonotylus coelestialium, Stenotus rubrovittatus,* and various mirid species occur throughout Japan. When grains are badly injured they ripen poorly and develop colored spots called 'peck'. Pecky rice has become serious since 1970 when overproduction of rice became a problem. Since 2000, RSBs have spread to 30-50% of rice growing area of ca. 1.7 million ha. Recent warm winters, hot summers, and an abundance of grassy weeds has accelerated population growth of RSBs. Economic thresholds (ET) have been developed based on field studies of natural populations. ETs of 0.8 adults per 5 sweeps with a 36-cm diameter net during heading and milk stages for *L. chinensis,* or much less for *T. coelestialium,* have been established. A double-strategy of weed control and insecticide applications is recommended for IPM of RSBs. When ETs are exceeded insecticides are applied 1-2 after heading at 10 days intervals, either by ground spray or by aircraft.

Management of *Lygus* spp. (Hemiptera: Miridae) on buckwheat in Manitoba, Canada

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In Manitoba, large numbers of *Lygus* bugs have been observed on buckwheat, *Fagopyrum esculentum*, particularly near the end of the growing season. However, there was no information about the effects of these bugs on yield. In 2002 and 2003, the species composition, seasonal pattern, economic implications, and control of *Lygus* bugs in buckwheat were investigated. *Lygus lineolaris* dominated the insect assemblage in buckwheat. A small number of adult bugs arrived in the buckwheat in late July and a generation of nymphs developed in the following month. In early September there were many more adults than had developed as nymphs, presumably indicating migration from early-maturing crops. An application of lambda cyhalothrin in late July suppressed nymphs, and resulted in yield weights averaging 59% higher than in unprotected crops. Insecticidal control of adult bugs in September did not significantly affect yield. *Lygus* bug infestations decreased yield weight and seed number/m^2, and increased the percentage of malformed seeds. In a trial of *Beauveria bassiana* against adults in September, seed yield and fungal infection of bugs were significantly greater in treated plots, although the number of bugs in treated and control plots did not differ.

Landscape structural design and vegetable pest management

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Biodiversity is one aspect of spatial pattern that is an important component of agroecosystem health. Although typically considered in terms of rare or endangered organisms, crops themselves can be part of the biodiversity that contributes to agroecosystem health. In fresh vegetable production, the landscape typically is composed of diverse mixtures of crops, each anchoring their own unique food web. The spatial scale and arrangement of vegetable plantings can affect the flow of insects, both pests and beneficial, across the landscape with consequences for insect control. Geostatistical analysis of two different insects suggests that favorable crops decelerate movement of highly mobile insect pests, whereas less favorable crops accelerate their movement within and perhaps out of production areas. Detailed spatially explicit simulation of aster yellows epidemics indicates that highly mobile insect vectors of plant disease are less likely to sustain epidemics if susceptible crops are adequately spaced. Research at two different spatial scales, within and between fields, demonstrates that the diversity within and between vegetable plantings can lead to bottom-up effects that sustain populations of biological control agents of insect pests. Spatial pattern statistics for crop arrangement might be used to design beneficial effects and avoid detrimental effects.

Evaluation of corn hybrids for tolerance to corn rootworm (*Diabrotica virgifera virgifera* LeConte) larval feeding

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The western corn rootworm (*Diabrotica virgifera virgifera* LeConte (WCR)) is a major pest of corn. The pest arrived in Croatia in 1995, and today over 250.000 ha are infested. Insecticides are regularly used to control WCR, but the cost is high, can pose environmental risks, and may become ineffective due to resistance. Growing corn that is resistant to corn rootworms would be a valuable alternative to insecticides. Nine Croatian (Institute of Agriculture, Osijek) and two Pioneer Hi-Bred Int. Inc. (Johnston, Iowa, USA) commercial corn hybrids were evaluated for WCR resistance at two locations in 2001, 2002, and 2003 year. One location was in east Croatia (Osijek) and one was in the USA (Iowa). The hybrids' tolerance to larval feeding was rated using root injury, root regrowth and root size. Root injury was rated using the Node-Injury Scale 0-3 and evaluation of root size and root regrowth were done by scale 1-6. In an analysis of variance combined across locations, root injury didn't show any significantly differences among hybrids, but root regrowth and root size were significantly different.

Comparative effects of *Leptocorisa oratorius* (F.) on the yield, grain quality, and seed germination rates of "super rice" and three other rice lines

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We studied the effect of *Leptocorisa oratorius* (Fabricius) on the yield, grain quality, and seed viability of four rice (*Oryza sativa*) cultivars. Three of the cultivars, C2, IR64, and PSBRc20 were released in the Philippines. The fourth variety, IR72164-201-1 is an unreleased experimental line of an *O. sativa japonica* X *O. sativa indica* cross, known in the popular press as "super rice". Each variety was exposed to four infestation densities for 21 days. *L. oratorius* feeding produced unfilled and partially filled grains, resulting in a negative correlation of yield to rice bug density. Germination rates were negatively correlated with rice bug densities. The percentage of discolored grains was positively correlated with *L. oratorius* density on all cultivars. Overall, PSBRc20 and IR64 had higher yields, less damaged grain, and higher germination rates than IR72164-201-1 and C2 under high rice bug infestations; suggesting the possibility of host plant resistance to rice bugs. The economic injury levels (EIL) currently used for rice bug management are based solely on yield loss estimates. The results of this study suggest that EIL for rice bugs should be revised to take into account reductions in grain quality and seed germination rates in addition to yield loss.

Preharvest field control as part of systems approaches to quarantine security

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National and international trade is regulated, in part, by insect quarantine issues that require knowledge of pests that are present or likely to be present in commodities as well as adequate assurances that the "risk" of introduction will meet requirements of the importing country. The development of systems approaches to quarantine security has more recently evolved to take into consideration the impact of preharvest pest control measures that reduce the pest population and therefore the risk that commodities that undergo further inspection or postharvest quarantine treatments are infested. Systems approaches require knowledge of the pest biology in the field, seasonal host phenology, efficacy of specific treatments and the impacts of such on the efficacy of potential quarantine treatments. For fruit flies, the polyphagus nature of many fruit fly species, alternate hosts present in tropical agroecosystems, year-round growing season and internal feeding of immature stages requires particular attention to the preharvest field situation. Areawide integrated pest management systems for fruit fly control have been implemented in many locations and have been shown to be successful in significantly lowering fruit fly populations. Other examples of preharvest methods that could affect postharvest quarantine issues include host prevalence, in-field sanitation and area freedom. Examples of these methodologies and how each may impact pest risk are discussed.

Japanese oak wilt as a newly emerged forest pest in Japan: why does a symbiotic ambrosia fungus kill host trees?

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Japanese oak wilt (JOW) has been known since the 1930s, but in the last fifteen years epidemics have intensified and spread to the islands' western coastal areas. The symbiotic ambrosia fungus *Raffaelea quercivora* is the causal agent of JOW, and is vectored by *Platypus quercivorus*. This is the first example of an ambrosia beetle fungus that kills vigorous trees. Mortality of *Quercus crispula* was approximately 40% but much lower for associated species of Fagaceae, even though each species had a similar number of beetle attacks. It is likely that other oaks resistant to the fungus evolved under a stable relationship between the tree, fungus and beetle during a long evolutionary process. *Quercus crispula* was probably not part of this coevolution. *Platypus quercivorus* showed the least preference for *Q. crispula*, yet exhibited highest reproductive success in this species. Therefore, *P. quercivorus* could spread more rapidly in stands with a high composition of *Q. crispula*. *Platypus quercivorus* can reproduce only one year on each *Q. crispula* tree. The relationship among *Q. crispula* – *R. quercivora* – *P. quercivorus* seems evolutionary unstable. JOW seems to be an invasive pest of *Q. crispula*. Concentric patterns of JOW spread also support this hypothesis.

Inducing resistance as a management tool

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Induced resistance involving vaccines and drugs that affect the immune response is widely used in western medicine. Similar techniques have been applied recently to manage plant pathogens and hold considerable promise against pest insects. Plants can be successfully vaccinated against herbivores by inoculating them with avirulent species, although these techniques are knowledge intensive and costly. Resistance may also be induced using cultural manipulations or by applying chemical elicitors. These methods do not directly kill the pests but enhance the plant's own defenses. Exogenous applications of methyl jasmonate have been found to induce resistance in many crop species against many insect pests under agricultural conditions. Other chemical elicitors have been identified that are more powerful than naturally occurring methyl jasmonate. Based on the lessons learned by plant pathologists, commercially successful elicitors will likely cause multigenic resistance against a broad spectrum of insects in many diverse crops.

Are island forests vulnerable to invasive defoliators?

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It has become axiomatic to represent oceanic islands as fragile ecosystems, which, because of their evolution in isolation, are particularly susceptible to loss of endemic species. It is patently obvious that simple stable ecosystems do exist (eg bogs, boreal forests and oceanic islands) and remain stable if left undisturbed by man. Risk assessments of the threat posed by invasive Lymantriids to the New Zealand flora, indicate that despite the accepted paradigms, New Zealand's flora is largely resistant to these exotic defoliators. As an explanation of this apparent anomaly, the Island Resource Allocation (*IRA*) hypothesis was developed which states that 'the palatability of plants to invertebrate herbivores is proportional to the geographic range of the plant'. The basis for the *IRA* hypothesis is a redefinition of the fundamental ecological principle of the species: area relationship. Species diversity increases with area, but the relationship is more pronounced at higher trophic levels. The redefined species: area relationship means that islands, or similarly geographically constrained ecosystems, which support lower biodiversity, have fewer trophic levels, and consequently have a lesser regulation of herbivores by natural enemies. The *IRA* hypothesis argues that island ecosystem stability is achieved through bottom-up processes, such as plant defences.

Silicon enhances resistance of sugarcane to African stalk borer *Eldana saccharina* Walker (Lepidoptera: Pyralidae)

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Soil amendments with silicon (Si) can increase crop resistance to insect pests, including that of South African sugarcane to its major pest, *Eldana saccharina*. This study examined the effect of six sources of Si, incorporated into the growing medium at low (T_1) and high (T_2) levels (controls = no Si), on resistance to *E. saccharina* of seven sugarcane varieties in a pot and a field trial. In pots (4 varieties, 4 Si sources), treatment T_1 (1200 kg/ha pure Si) reduced mean borer damage and borer survival by 34.4% and 30.8%, respectively, in the two susceptible varieties and by 25.7% and 13.2%, respectively, in the two resistant varieties. Effects were significant only at T_2. In the field (3 varieties, 2 Si sources), mean borer damage was reduced by 20.5% in the two susceptible varieties at T_2 (480 kg/ha pure Si) and by 21.5% at T_1 (960 kg/ha pure Si); in the resistant variety damage was reduced by 10.6% (T_1) and 16.7% (T_2). Borer survival was reduced only in the susceptible variety (T_1: by 14.3%; T_2: by 26.7%). The results support the argument that Si amendments in Si-deficient soils, can substantially reduce sugarcane stalk damage and yield loss due to *E. saccharina*, especially in susceptible varieties.

Salt mixture - an IPM option for managing sucking pests in cotton

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Green vegetable bug (GVB), *Nezara viridula* (L.) and green and brown mirids, *Creontiades spp.* have emerged as important mid to late season sucking pests in cotton in Australia. The insecticides currently registered for the control of these pests are mostly non-selective and are highly disruptive to a wide range of beneficial species. To increase selectivity of the existing chemicals we are using common salt (NaCl) to mix with reduced rates of chemicals. The mixture provided good control of GVB and mirids. They also decreased the impact of treatments on beneficial insects and spiders. Trials were carried out to evaluate Regent® (fipronil) plus salt against mirids and GVB, Steward® (indoxacarb) plus salt against mirids and Affirm® (emamectin) plus salt against GVB. Trials were also conducted to determine the effect of salt rate on the mixture and to find out the reason for salt mixture working. The results indicate that salt mixture increased mortality by 20 to 40% compared to reduced rates of chemicals alone and were as effective as full rates of chemicals. The results also indicate that effectiveness of the mixture did not vary with salt rate and salt mixture encouraged insects to probe 30 – 35% more compared to chemical treatment alone. The implication of this finding for management of GVB and mirids in the context of cotton IPM will be discussed.

Synthetic hexaploid wheat: increasing diversity for resistance to Russian wheat aphid and greenbug

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Fifty-eight different *Triticum dicoccum* x *Aegilops tauschii* synthetic hexaploid wheats were evaluated for resistance to Russian wheat aphid (RWA) (*Diuraphis noxia*) and greenbug (*Schizaphis graminum*) under field and greenhouse conditions. In the field, plants were grown in one-meter long, paired plots, one plot infested with RWA while adjacent plot treated with insecticide. Resistance observed in the *T. dicoccum* parents was expressed in the synthetics, although variation occurred. All *Ae. tauschii* parents were susceptible indicating that any RWA resistance present in the synthetics must be located on the A and/or B-genomes, whereas all other previous located RWA resistance genes are located on the D-genome. Greenhouse evaluation for greenbug resistance revealed resistance in 9 out of 15 randomly selected *Ae. tauschii* parents, while all *T. dicoccum* parents were susceptible. Resistance was expressed in 75% of the synthetics derived form resistant *Ae. tauschii* parents. However, both positive and negative epistatic interactions was also observed. These results show that: 1) synthetic hexaploid wheat can act as "genetic bridge" for RWA and greenbug resistance, 2) novel genetic diversity for RWA resistance can be derived from *T. dicoccum*, 3) *Ae. tauschii* is an important source of greenbug resistance, 4) epistatic interactions can occur in synthetic hexaploids.

Assessing the susceptibility of plantation *Pinus* spp. in Queensland to pine wilt nematodes

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In 2001 an incursion of *Monochamus alternatus* (Coleoptera: Cerambycidae) was detected in Brisbane in pallets imported from China. This beetle is the primary vector of pine wilt nematode (*Bursaphelenchus xylophilus*) a devastating pest of *Pinus* spp. in northeast Asia (Japan, China and Taiwan). The susceptibility of the major pine species grown in Queensland (predominantly *Pinus caribaea* var. *hondurensis* and its hybrids with *Pinus elliottii* var. *elliottii*) to this nematode was unknown. To test relative susceptibilities of both the parent species and the six best performing hybrids, one-year old seedlings were sent to Japan in 2002 and 2003 and planted out in a nursery at the Forestry and Forest Products Research Institute, Tsukuba. These were later inoculated with either distilled water, *B. xylophilus* or *B. mucronatus,* as dispersal stage adults. *B. mucronatus* is endemic to Eurasia and is not pathological to *Pinus* species there, but its pathogenicity against southern USA and Caribbean pines was unknown. Additionally, stems of all eight pines were presented to captive adult *M. alternatus* beetles to test for maturation feeding acceptability. The results of this research will be commented on in relation to risk management and proactive breeding programs for exotic pest resistance in Queensland.

Susceptibility of *Eucalyptus* taxa to attack by giant wood moths (*Endoxyla cinerea*) and longicorn beetles (*Phoracantha* spp.) in Queensland

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Since 1997 more than 500 hectares of trials of eucalypt taxa have been established in Queensland to evaluate their potential as commercial plantation species. Plantings of eucalypt species in northern New South Wales and southeast Queensland between the 1960s and the early 1990s confirmed that a number of stem boring insects were potentially important pests limiting plantation productivity. The giant wood moth *Endoxyla cinerea* and longicorn beetles in the genus *Phoracantha* were the most important of these. To evaluate the susceptibility of eucalypt taxa to these stem borers, 25 eucalypt taxa were assessed at 19 trial sites across southeast and north Queensland by visual inspection of external damage to 2.5 to 3 y.o. trees. Priority solid timber species planted in southeast Queensland, *C. citriodora* ssp. *variegata*, *E. cloeziana* and *E. argophloia*, generally showed low incidences of attack. In contrast, the three main species being grown in Queensland for pulp purposes, *E. grandis*, *E. dunnii* and an *E. grandis* x *E. camaldulensis* hybrid all showed moderate to high rates of attack. Our results are commented on in relation to the future management of these insects and the expansion of the hardwood plantation industry in Queensland.

Thiamethoxam as a drench application for the control of scale insects and mealy bugs on mangoes in South Africa

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The control of the scale insect ***Aulacaspis tubercularis*** & various mealy bug species on mangoes in South Africa has become problematic due to the withdrawal of several products. To address this problem, imidacloprid, acetamiprid and thiamethoxam (neo-nicotinoids) were tested over three seasons in large field trials. During the 2001/02 season all three products produced promising results with thiamethoxam being the most efficacious. In 2002/03 dosage verification trials were conducted on the three products. The level of insect control achieved by imidacloprid and acetamiprid was not cost effective. However, thiamethoxam proved to be cost effective. Further development trials with thiamethoxam commenced in 2003/04 in three mango growing areas of South Africa. In all three regions it was shown that the placement of the drench application was important. Application at the point of drip irrigation was most effective, resulting in approximately 90% of the crop being of export quality and free of insect pests. The untreated control plots yielded 20% - 50% export quality fruit.

Integrating crop resistance and biocontrol for sustainable protection against wheat midge

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An antibiotic resistance gene *Sm*1 will be deployed against larvae of the wheat midge, *Sitodiplosis mosellana* (Géhin) (Diptera: Cecidomyiidae), an important pest of wheat in the Northern Hemisphere. This resistance kills over 95% of larvae in the field, but its efficacy is likely to decline as virulence to *Sm*1evolves. Oviposition deterrence, probably conferred by two genes, has also been identified, but no other antibiosis genes are available. Hymenopteran egg parasitoids and insecticide applications now contribute to pest management, although imperfectly. An interspersed refuge of 5-10% susceptible plants within the resistant crop could slow or prevent the evolution of virulence and preserve parasitoid populations, while maintaining damage below economic thresholds. The refuge is greatly enhanced by pyramiding *Sm*1 with oviposition deterrence. This integrated resistance strategy has been assessed in multi-year field trials of the refuge, by investigating the prevalence of virulence in the midge population, and by computer simulation. The critical features of the insect-plant interaction that require an interspersed refuge and contribute to the success of the strategy are: a sedentary feeding stage, sedentary male midges, post-mating dispersal by female midges, dispersed oviposition which stabilizes local sex ratio, and density-independent searching by the parasitoid.

Host tree resistance to insects in changing forest communities

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Tree resistance to insects can condition the nature and establishment of invasive insect and introduced tree species, as well as the moving direction of the invaders, in various ways. Effects of resistance of indigenous or introduced tree species on invasion processes of exotic or indigenous insect species, respectively, are first considered. Factors involved can be: - botanical diversity; - phenological coincidence and host plant distribution; - resemblance between defence mechanisms of new hosts and those of the original host species or resemblance of the introduced tree species with local species; - capacity of native or introduced trees to adapt to new aggressors (role of previously encountered selective pressures); - capacity of invasive insects to adapt to resistance of new hosts (role of selective pressures existing in the original habitat and role of the presence of original host trees). Climate change can also affect resistance of indigenous trees, then influencing invasion processes of insect populations. The effects of introducing exotic tree or insect species on resistance of indigenous trees are then discussed. These factors can act through: - modification of forest biodiversity, tree competition, vigour and growth; modifications of microclimatic and soil conditions; modification of tree attraction; interactions between insect species.

Compatibility of biopesticides and reduced-risk products with biological control agents of *Plutella xylostella* in cabbage: problems and perspectives

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Biopesticides, including Bt-products and azadirachtin-based insecticides, have been extensively used for management of *Plutella xylostella* and other lepidopteran pests in crucifer vegetables in the US. Spinosad and indoxacarb are two relatively newer insecticides with distinct modes of action and chemistries from other conventional insecticides, and have been considered as reduced-risk insecticides by the USEPA. However, our laboratory bioassays (contact and ingestion) indicate that both spinosad and indoxacarb can be destructive or exhibit significant effects to pupae and adults of *Diadegma insulare* and *Cotesia plutellae,* parasitoids of *P. xylostella*. In contrast, all Bt-products and azadirachtin-insecticides had no or little effects to the two parasitoid species. Our field studies also reveal that parasitism of *P. xylostella* by *D. insulare* on cabbage in the treatments of Bt- and azadirachtin-insecticides are generally greater than in the treatments of spinosad, indoxacarb, and l-cyhalothrin, indicating that the latter three had negative effects to the parasitoids and other natural enemies. We conclude that all Bt- and azadirachtin-insecticides are compatible with the parasitoids in IPM programs against *P. xylostella* in crucifer vegetables, whereas spinosad and indoxacarb need to be used with caution when *D. insulare* and *C. plutellae* are dominant in the cabbage fields.

Biological impact of the exotic diamondback moth parasitoid *Diadegma semiclausum* (Hellen) in Kenya

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Pre and post-release studies were conducted in farmer-managed crucifer fields to assess the biological impact of the parasitoid. Fortnightly, diamondback moth (DBM) populations and damage were assessed in fifteen farmers' fields. Larvae and pupae were collected to assess parasitism and 7 and 12 months after release, exclusion experiments were conducted to assess parasitoid contribution to mortality. Before release, populations peaks were 15.8 DBM/plant (Oct.-Nov.) and 7.9/plant (Jan.-Feb.), average annual population was 5.8 and the damage score was 1.9.Most farmers applied insecticide weekly. The first year after release, peaks declined to 5.5 and 1.9/plant, annual average was 3.9. The damage score remained similar but farmers stopped pesticide application. Parasitism in exclusion experiments increased from 18.0% to 65.0% at Werugha and from 9.2% to 18.1% at Limuru, 7 and 12 months after release. Data from exclusion experiments and field collections indicate displacement of indigenous parasitoids. In field simulation experiments with different parasitoid densities, parasitoids increased unexplained loss of larvae. Most larvae collected off-plant (68-75%) were parasitised. We conclude that field parasitation rates measure less than half of the killing power of the parasitoid. All results indicate high impact of the parasitoid and justify releases all over East African highlands.

Development of an IPM program for cotton production in Arkansas, 1997-2003

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There have been numerous IPM projects and programs in Arkansas over the years, many achieving great successes in reduction of pesticide usage within particular crops and increased economic benefits to growers. In response to the National Initiative and in an effort to increase and better coordinate actual farm implementation, a county-based IPM program was initiated in 1997. This program offers financial and specialized support to participating counties. Counties submit project proposals, which are screened and approved (or not) by an IPM Program Committee. Documentation of program efforts and results is required. Arkansas farmers have traditionally used many IPM methods but largely in an unfocused way. Similarly, researchers and extension personnel have developed pest management recommendations using many biological, cultural, and chemical techniques but more often than not, have failed to integrate them into a single, cohesive system for growers to use. The need for IPM education is obvious today, with increasing challenges to crop production for Arkansas growers. In Arkansas, the success of implementing any IPM Program will rest logically and predominately on the County Extension Agents. This is as it should be, since local education efforts are generally the most successful in the short and long term. The county agent system has obvious close contact with the farmer, but also works closely with consultants and industry representatives at the local level. The results of this program have shown great success in the implementation of IPM in Arkansas agriculture.

Successful suppression of melon fly, *Bactrocera cucurbitae*, on Oahu and Maui, Hawaii

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The melon fly, *Bactrocera cucurbitae* is the most harmful pest of cucurbit and melon crops in Hawaii. Before the area wide program was implemented, growers reported losses 40-60 percent and occasional complete losses. We implemented a science-based integrated pest management (IPM) philosophy and a comprehensive grower education program. In addition to establishing a monitoring program, step-wise implementation of the following technologies was implemented. The tactics were: 1) field sanitation; 2) environmentally safe spinosad bait sprays; 3) male annihilation. Geographical positioning systems (GPS) and geographical information systems (GIS) tools were used to determine the impact of the IPM tactics on fruit fly densities throughout the implementation zones. As a result of the program, melon fly populations are at an all-time low. Peak male populations at the start of the program in 2000 were 100, and 135 flies/trap/day, respectively at Kula, Maui and Ewa, Oahu implementation zones. Fly population densities at each of the implementation zones were reduced to about 5 and 25 flies/trap/day, respectively. Melon fly percent infestation rates were reduced from >30 percent on Oahu to less than 5 percent at in 2003. Organophosphate usage as crop sprays for melon fly control was practically eliminated.

An evaluation of reduced risk pesticides for control of lepidopterous pests of cole crops

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The major pests of cole crops are three caterpillars; diamondback moth, *Plutella xylostella* (L.) (DBM), imported cabbageworm, *Pieris rapae* (L.), (ICW), and cabbage looper, *Trichopulsia ni* (Hubner). These insects are all pests in the larval stage, as eggs are laid on the surface of the leaves and the caterpillars feed on the foliage as they develop. They are generally viewed as a single caterpillar complex and controlled as such. Traditionally broad-spectrum insecticides have been used to control these pests. However in recent years there is growing concern of food safety, resistance, environmental protection, and conservation of the natural enemy guild. Therefore, the pressure to find new control methods that are safer for humans and the environment and that maximize the efficacy of natural enemies for control of these caterpillars is increasing. This study looks at *Bacillus thuringiensis* (Bt) and specifically compares Dipel DF, Xentari and a combination of the two for control of these pests. In addition, the efficacy of EnTrust (a microbial product) is compared to the *Bt* sprays to get a good comparison of an array of reduced risk pesticides.

Evaluation of petroleum spray oils for integrated pest management program against cotton pests in Australian cotton agro-ecosystems

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Petroleum spray oils (PSOS) have been used to control a wide range of crop pests and are now an essential part of many integrated pest management (IPM) programs. We determined the efficacy of an nC27 based oil (Caltex Australia Canopy®) oil) against *Helicoverpa* spp. on cotton crops in the field and mesh house. The results of the study showed that application of 2% v/v of the PSO was efficacious against *Helicoverpa* spp. larvae, reduced oviposition by *Helicoverpa* adult moths and enhanced the efficacy of nuclear polyhedrosis virus (NPV) and *Bacillus thuringiensis* (Bt) against *Helicoverpa* larvae with no symptoms of phytotoxicity to cotton plants. A comparison of plants treated with PSO at various times showed that only 50% of tested females settled on plants 4-5 days after treatment (DAT) whereas none settled on plants 0-2 DAT. A solid phase micro-extraction (SPME) test to determine the effect of the PSO on volatiles released by the cotton plants showed that the quantity of volatiles released by the cotton plants treated with PSO was relatively lower than water treated plants. This indicates that the PSO sprays may be suppressing or masking the leaf surface volatiles of the cotton plants, thereby deterring oviposition of *H. armigera*. PSOs may have the potential to be integrated into IPM programs against cotton pests to minimise synthethetic insecticide use on cotton crops.

Reducing the threat of Russian wheat aphid on wheat and barley in Australia

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The Russian wheat aphid (RWA) *Diuraphis noxia* (Mordvilko) occurs in all major cereal-growing regions worldwide except Australia. As a preparedness strategy against RWA, GRDC and CSIRO funded research to develop RWA-resistant Australian germplasm. Research at the CSIRO laboratory in Montpellier, France investigated the performance of four RWA populations (United States (US), South Africa (SA), Hungary (HU) and France (FR)) on five wheat lines (PI 137739 (Dn1), PI 372129 (Dn4), Halt (Dn4), Arminda (S) and Wichita (S)) and one susceptible barley. Seedlings at the two-leaf stage were infested with aphids of each of the four populations. Three weeks later, several plant damage symptoms were recorded. Overall, HU significantly differed from the other three populations ($P<0.0001$) by causing the most damage on all plants, and there were no significant differences ($P>0.05$) among US, SA and FR populations. Comparisons among plants showed that Dn1 exhibited the best resistance against the aphids. Dn4 was as good as Dn1 when subjected to three populations, FR, SA, and US. Research is ongoing to test additional Dn and Dbn genes, and will hopefully incorporate other virulent RWA clones. These and other findings will provide a basis for crossing effective Dn genes into Australian germplasm.

Population dynamics and efficacy of ULV aerial application of diflubenzuron against the cedar web spinning sawfly, *Cephalcia tannourinensis* Chevin, in Lebanon.

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The cedar web-spinning sawfly *Cephalcia tannourinensis* Chevin is a pest that has been causing serious damage to cedar forest of Lebanon since 1990. The populations of the pest is composed of individuals characterized by a different length of the life cycle, annual and multi years individuals. The two parts of the population differ as to the distribution of their larvae (prepupae) in the soil; long term diapausing larvae being deeply distributed in the soil. The ULV air-application technology was conducted from 1999 until 2002 with the insect growth regulator diflubenzuron. Evaluation methods were done by sampling prepupae in the soil, catching adults with yellow sticky traps and direct observation of larvae on branches. The results ranged from 60 to 85% mortality of the larval stages present on randomly selected branches depending on years. One-year diapausing prepupae population decreased up to 70% in treated areas compared to 52% decrease with multiple years diapausing prepupae. A significant linear relationship between estimates of adult catches and number of prepupae ready to emerge was found. The variability in the diapause length makes it difficult to predict future attacks and the phenology.

Real-time prediction of planthopper migrations in East Asia by three-dimensional computer simulations

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The white-backed planthopper and the brown planthopper are insect pests of rice and migrate from southern China to Japan in the rainy season. In order to develop a high-precision migration prediction system, an advanced numerical weather prediction model, MM5, and a particle random-walk model, GEARN, were employed. The real-time meteorological data are supplied online to MM5 which can forecast three-dimensional atmospheric fields at one-hour intervals. GEARN calculates the movement of a number of planthoppers and predicts relative aerial density. The forecasting results are provided as geographical maps on the Internet. The changes of the relative aerial density show the timing and area of migrations over the next two days. An evaluation was conducted using daily catch data collected in the south-western Japan in 2003. The result showed the prediction system worked as required throughout the migration season and the prediction quality was comparable to that of rainfall forecast by the Japanese Meteorological Agency.

Changing the mix: new rules in regulating herbivore populations

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Populations of herbivores in native or endemic environments are regulated by a broad range of factors that can include abiotic conditions and both top-down and bottom-up factors. It is possible to characterize the relative importance of each factor and the conditions where the balance among regulating factors can change. However, when those herbivores become established in novel environments, the factors regulating the populations and the balance among those forces can be astically altered. The host plants are growing under very different conditions, often in association with very different communities with different composition and structure. These differences can change the bottom-up regulation of the herbivore populations. The herbivore may also be part of a very different community, either more simplified with fewer potential competitors, or more complex. The community of natural enemies may also be simplified or more complex. Consequently, the impact of top-down regulation on the population may be very different from what is observed in the native range. The differences in the impact of individual components that contribute to population regulation need to be carefully assessed in the context of the novel environment, host tree community, other herbivores, and complex of natural enemies.

Aphids associated with papaya plants in Puerto Rico

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In Puerto Rico, papaya (*Carica papaya* L) is the most extensively grown fruit. The crop is severely affected by the papaya ringspot virus (PRV), causing severe economic losses. With all the interest in the commercialization of papaya in Puerto Rico, there is a paucity of information on the vectors of the PRV in the island. Field trials were conducted at the Agricultural Experimental Station, University of Puerto Rico, Isabela and Corozal to identify aphids associated with papaya. Ten species of aphids were identified: *Toxoptera citricida* (Kirkardy), *Toxoptera aurantii* (Boyer de Fonscolombe), *Pentalonia nigronervosa* Coquerel, *Tetraneura nigriabdominalis* (Sasaki), *Uroleucon ambrosiae* (Thomas), *Aphis spiraecola* Patch, *Aphis gossypii* Glover, *Aphis craccivora* Koch, *Aphis illinoisensis* Shimer and *Picturaphis sp.* All species were present at Corozal, while only six species were collected from Isabela. The difference in species composition between localities is associated with the crops present in each location. Plantains and papaya are common in both localities; however commercial papaya plantings are common in Isabela, while plantains and citrus are predominat crops in Corozal. The presence of weeds in fields could be an important factor. The plantings at Corozal were almost weed free, while at Isabela weeds were common and abundant between papaya rows.

Biological control of *Pulvinaria urbicola* (Cockerell) in a *Pisonia grandis* forest on North East Herald Cay in the Coral Sea

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During 1993-2000, *Pulvinaria urbicola* (Cockerell) completely destroyed a 16 ha *Pisonia grandis* forest on Coringa South West Islet, 400 km off the coast of Queensland in the Coral Sea. Surveys on nearby North East Herald Cay, with a similar area of Pisonia, showed a worrying increase in scale infestation in December 2000. A survey in March 2001 revealed no natural enemies and five months later, the mealybug ladybird *Cryptolaemus montrouzieri* (Mulsant), and three parasitioids, *Coccophagus ceroplastae* (Howard), *Euryischomyia flavithorax* (Girault), and *Metaphycus luteolus* (Timberlake), were released. *C. montrouzieri* multiplied and within six months reduced the scale population by 60%. Percent infested leaves dropped from 25% to 8%, and the scale average per cm of midrib from 0.34 to 0.18. One year after release of the natural enemies, the scale average dropped to 0.03. Two years after release, only 0.01% of leaves were infested. The ants, *Tetramorium* sp., and *Monomorium* sp., feeding on the scale honeydew, declined by 99%. All three parasitoids established, and parasitism was evident on 30% of infested leaves after nine months. The parasitoids, particularly *C. ceroplastae*, are expected to play a important role in maintaining the scale population at its current low level.

Biological control of cotton aphids, *Aphis gossypii*, **in cotton: effectiveness of** *Hippodamia convergens* **and** *Chrysoperla carnea*

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"The predation rates of *Hippodamia convergens* (adult and larva) and *Chrysoperla carnea* were quantified at fixed prey density (100 aphids/leaf) in the laboratory. *Hippodamia convergens* adult showed the highest rate of consumption (91 aphids/12 h) followed by *C. carnea* (75 aphids/12 h), *H. convergens* medium larva (56 aphids/12 h), and *H. convergens* small larva (13 aphids/12 h). Predation rates were also examined temporally for variable prey densities. Laboratory prey suppression trial and field cage study showed that *H. convergens* adult can suppress *Aphis gossypii* populations more efficiently at lower prey density and the suppression rate decreased when prey populations exceeded 100 aphids per leaf. In a field cage study, one *H. convergens* adult per plant released at the prey density of one aphid per leaf kept the aphid population below economic threshold for the entire growing season. Three *H. convergens* adults per plant released at 25 aphids per leaf or six adults per plant released at 50 aphids per leaf could not prevent aphid populations from exceeding the economic threshold. Field experiments showed strongly positive temporal and spatial distributions of *H. convergens* with that of *A. gossypii*, suggesting a significant potential of *H. convergens* for *A. gossypii* suppression in cotton.
"

Development of banana entomology in north Queensland, Australia: progress towards an IPM system.

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Over 80% of the Australian banana production is concentrated on a narrow strip of Queensland's tropical coastline from Cardwell in the south to Innisfail in the north. This paper presents the changes in pesticide use and the new developments that have occurred in application methodology against the five major pests of commercial bananas in north Queensland. Other minor pests and quarantine threats are also discussed. Chlorinated hydrocarbons had been used since the 1960s primarily for control of the two major pests, the banana scab moth, *Nacoleia octasema* and the banana weevil borer, *Cosmopolites sordidus.* Legislative bans placed on these in 1985 resulted in a switch to organophosphates and other less disruptive and shorter residual insecticides. The universal adoption of targeted injection into the emerging bunch has resulted in a 95% reduction in pesticide use. Controlled release pesticide technology and, more recently, biopesticides, are further reducing off-target impacts for the control of bunch pests. The changes in pesticides and development of the new application technologies are discussed in relation to IPM.

Unraveling the etiology of aphid-elicited desistance to improve aphid resistance in wheat

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The etiology of aphid-elicited desistance on wheat (*Triticum aestivum*) was studied using 'Tugela' and three near-isogenic lines and two cereal aphids (chlorosis-eliciting *Diuraphis noxia* and non-chlorosis-eliciting *Rhopalosiphum padi*). The mechanisms of aphid-elicited chlorosis will be discussed in the context of photosynthetic pigments (chlorophylls *a* and *b* and carotenoids) and chlorophyll degradation enzyme (chlorophylase, oxidative bleaching, and Mg-decheltase) activities. Concentrations of chlorophyll *a* and *b* and carotenoids were lower in *D. noxia*-infested wheat lines compared with *R. padi*-infested plants. Chlorophyllase activities were higher in *R. padi*-infested plants and lower in *D. noxia*-infested plants, while reverse mg-decheltase activities were observed. *Diuraphis noxia*-induced chlorosis was most likely driven by unbalanced chlorophyll biosynthesis and catabolism processes.

Targeted control of *Myzus persicae* (Sulzer) in North American seed potato production

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Mid-summer colonization of seed potato fields in the northern plains of North America by *Myzus persicae* (Sulzer), the key PLRV vector, is concentrated on field edges, particularly at the crop interface with fallow or owned out areas, but especially along headlands. This colonization behavior combined with the fact that apterae move <2 m per day within the field provides a spatial and temporal window for site-specific pest management. In 2003, large-scale on farm experiments (25 seed potato fields, each >20 acres) evaluated the effectiveness of targeting spray treatments for *M. persicae* on field edges. When *M. persicae* flight activity spiked (~July 30), an 18 m wide spray swath of methamidophos insecticide was applied by airplane to field edges abutting headlands. Pretreatment densities of *M. persicae* were 16 times higher on field edges than mid-field. Three days post-treatment, *M. persicae* densities on field edges were reduced 92% from pretreatment levels. Seven days post-treatment, *M. persicae* densities in untreated field interiors were still not significantly different than in sprayed edges. Treating only 94 of 1804 acres saved 85% in control costs ($23.85 per acre, application included) compared to treating all acres.

Aphids and blueberry scorch virus – a multifaceted problem

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Ericaphis fimbriata (Richards) (Homoptera: Aphididae) is the predominant aphid on blueberry, *Vaccinium corymbosum* L. (Ericaceae), in British Columbia, Canada, and a vector of *Blueberry scorch virus* (BlScV). BlScV may reduce yield by more than 85% depending on virus strain and plant cultivar. Virus transmission is non-persistent so resident aphids on blueberry, as well as winged morphs of other species that frequent blueberry fields, may transmit the virus. Our studies suggest that local spread of BlScV is facilitated by the life history and behaviour of *E. fimbriata*: egg hatch is timed with bud break; a high proportion of alatae are produced in the second generation – production is dependent on plant tissue type and local aphid density; populations grow exponentially through the spring; and apterae move frequently. Population simulations based on our life-history data for *E. fimbriata* show that about 45% of population decline during summer is caused by reduced plant quality. Growers normally apply chemical aphid controls in June. Our studies suggest that the appropriate timing for an IPM program is April, before bloom.

Resistance to hunting billbug, *Sphenophorus venatus vestitus* **among** *Zoysia*

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Hunting billbugs (*Sphenophorus venatus vestitus*) damage *Zoysia* and *Cynodon* but it is often misdiagnosed as drought, insect or disease damage. Plants of nine *Zoysia* cultivars were established and adult billbugs were confined on them in field cages in a paired cage split-plot experiment with four reps in June 2000, so infested and uninfested paired plants could be compared. Billbug damage was measured in late Sept. 2000. Shoot and leaf necrosis was evaluated by ranking each test plant on a scale of 1 to 9. Total rhizome and stolon length were measured and total plant biomass was oven ied and weighed. Resistance was identified in the four cultivars of *Z. matrella* ('Diamond', 'Cavalier', 'Royal' and 'Zorro') while the five *Z. japonica* cultivars ('Crowne', 'De Anza', 'El Toro', 'Meyer,' 'Palisades') were susceptible. Meyer, the industry standard, has now been documented as highly susceptible to zoysiagrass mite, fall armyworm, tropical sod webworm, tawny mole cricket, differential grasshopper and hunting billbug. The data provides the first report of *Zoysia* resistance to the hunting billbug.

Management of thrips in Florida citrus

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Several thrips species are known to be pests of citrus in one or more parts of the world. Depending on the species of thrips, feeding injury may occur on the twigs, leaves, flowers or fruit of citrus. In Florida, *Frankliniella bispinosa* (Morgan) is the prevalent thrips species feeding on the flowers of citrus. Infestation of citrus flowers with high numbers of *F. bispinosa* can lead to reduced fruit set but is not known to cause damage to developing fruit after petal fall. Another thrips species not associated with Florida citrus, *Scirtothrips citri* (Moulton), feeds on the rind of developing fruit causing ring scarring that results in the fruit receiving a lower grade at the packing house. Within recent years, localized problems on 'Murcott' tangors in Florida have shown somewhat similar rind blemish damage. Some growers have attempted insecticide applications during bloom to reduce this injury. In this paper, we provide a summary of thrips management in Florida citrus and preliminary results of a study examining the cause of ring scarring on 'Murcott' tangors in Florida.

Best management practices to control the cotton aphid, *Aphis gossypii* Glover, in strawberry grown under protected culture

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In the greenhouse, the cotton or melon aphid, *Aphis gossypii* Glover, is one of the most important pests worldwide. In order to determine the susceptibility of different strawberry cultivars, *Fragaria x ananassa* Duch to natural infestations of the cotton aphid, seven cultivars were grown and evaluated. Treatments were arranged in a randomized complete block with three replications per treatment (cultivar). No insecticides or fungicides were used at any time. Five releases of *Aphidius colemani* at a rate of 10 wasps per m^2 per month were made to control aphids. 'Banker plants' were introduced in the greenhouse to ensure the permanence of the parasitic wasps throughout the season. The effectiveness of the control was measure by selecting ten strawberry leaflets at random and by counting the total number of aphids and parasitized aphids ('mummies') per leaflet. Early releases of *A. colemani* can guarantee the effective control of the cotton aphid.

Cereal aphid expert system and the glance 'n go sequential sampling system for managing of greenbug (*Schizaphis graminum* Rondani) in winter wheat.

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An Expert System was developed to help wheat producers and agri-business essionals manage greenbug, *Schizaphis graminum* Rondani and other cereal aphids in winter wheat. The system has four components. Two components help the user identify common cereal aphids and their natural enemies. A third component allows the user to calculate a treatment threshold for greenbugs, based on control costs and grain yield potential. This component uses a yield loss model and a temperature-dependent development model for greenbug that links to a regional historical weather database. Once a treatment threshold is calculated, the user can select and print the appropriate presence/absence sequential sampling "*Glance 'n Go"* form to quickly and accurately determine if greenbug treatment thresholds have been exceeded in that field. The fourth component helps the user select the appropriate pesticide for corrective control of a greenbug outbreak using various criteria listed on the label. This system is designed to incorporate new research results as they become available. Future improvements will include incorporating the impact of aphids on forage yield and the inclusion of a presence/absence sampling tool for simultaneously assessing the potential impact of the aphid parasitoid, *Lysiphlebus testaceipes* on greenbug populations.

Successful mass-trapping in a bark beetle: effect estimation by 22-year time series

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Thaumetopoea pityocampa (Den. & Schiff.) (Lep.: Thaumetopoeidae) is one of the most destructive defoliators of natural and artificially established *Pinus* spp in the Mediterranean region. Infestation rates differ dramatically among species and tree individuals, suggesting female choice of hosts during egg-laying. We have identified two main factors affecting the host selection process: - Tree silhouette, suggested as a dominant factor in the litterature. In a rearing room we offered to moths several *Pinus nigra* trees, a preferred species, of different sizes. A significant positive correlation was found between the number of egg masses laid on each tree and both tree height and silhouette. - Volatiles of host plant may play an important role in habitat selection. We investigated the olfactory host recognition mechanisms of females by the gas chromatographic-electroantennographic technique (GC-EAD). We tested natural volatile blends and a synthetic mixture of monoterpenes present in pine needles. Repeatable responses were among others to minor compounds like *E*-ocimene, b-terpinol and neodihydrocarveol. In addition, the identification of suitable needles through contact sensilla on tarsi is evaluated as an additional input for the host choice by females.

Bemisia and *Liriomyza* complexes: implications for management on vegetables

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The whitefly *Bemisia tabaci* (Genn.) and the leafminers *Liriomyza huidobrensis* (Blanchard) and *Liriomyza trifolii* (Burgess) are globally distributed. *B. tabaci* has been a major pest in arid regions of the world for at least 70 years, particularly on agronomic crops. A worldwide outbreak of the pest that began in the late 1980s on vegetables and other crops was attributed to a new strain or even a new species, although this latter view is not accepted universally. Numerous strains have been identified since the outbreak using electrophoresis and RAPD PCR techniques. Differences in host suitability and insecticide resistance of some strains have been demonstrated. Throughout the late 1970s and 1980s, *L. trifolii* was a major pest on vegetables while *L. huidobrensis* was a minor pest on just a few crops. In the early 1990s, reports indicated that *L. huidobrensis* was becoming the dominant species in many areas, suddenly developing a much wider host range and tolerance to pesticides. Classical mating studies showed that the more destructive *L. huidobrensis* was genetically dominant, resulting in rapid spread through populations. Subsequent molecular studies have suggested the presence of cryptic *Liriomyza* species. Correct strain/species identification is important in determining appropriate biological and chemical control strategies.

Advances in wheat stem sawfly management

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Wheat stem sawflies (Hymenoptera: Cephidae) are key pests of wheat in the northern hemisphere. At least seven species of sawfly attack wheat in different parts of the world. Yield losses of more than 30% have been reported from North America, Europe, North Africa, and Asia. Current management practices include the use of resistant wheat cultivars, various cultural control practices, and biological control. Chemical control is not cost effective in North America and Europe, and is not widely practiced in other wheat production areas. We review the status and impact of wheat stems sawflies in different global regions, and current pest management alternatives. Several new sawfly resistant wheat cultivars have been released in the past few years. Recent research on cultural practices, particularly integrating tillage and biological control, is also highlighted. Finally, progress in finding and importing new exotic biological control agents for *Cephus cintus*, a pest species in western North America, is also summarized.

Getting help from the wild: exploitation of wild relatives of crops as source of novel genes for resistance to insect pests

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Identification and utilisation of germplasm with insect resistance is key for sustainable crop protection. Evaluation of germplasm for resistance to insects has revealed the presence of low to moderate levels of resistance against the key pests such as bollworms, pod borers, aphids, shoot flies, jassids, and white flies. Thus, progress in developing insect-resistant cultivars has been slow. Therefore, there is a need to find resistance genes in wild crop relatives. Resistance to specific pests in wild relatives has been found, as follows: *Heliothis/Helicoverpa* in cotton, pigeonpea, chickpea, and tomato; *Atherigona*, *Chilo*, and *Stenodiplosis* in sorghum; *Maruca* in cowpea; *Orseolia* and *Scirpophaga* in rice; *Amrasca*, *Aproaerema* and *Spodoptera* in groundnut; Hessian fly in wheat. Considerable information has also been generated on the physico-chemical mechanisms of resistance to the target insect species. The genes from wild relatives can be tapped through wide hybridisation, marker assisted selection, and sequencing and cloning of resistance genes for use in genetic transformation to increase the levels and diversify the bases of resistance to insect pests.

The status and challenges of insecticide resistance in vegetables

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Insecticide resistance represents an important challenge to the continued production of high quality vegetables for local and international markets. Some insect species have demonstrated an amazing propensity to develop resistance to synthetic, organic and microbial insecticides. A current database lists a total of 543 cases of arthropods that have developed resistance, and this list contains many important species attacking vegetable crops. Elevated crop losses can be incurred even before resistance is detected and continued reliance on the pesticide will often result in increased dosages or applications, thus leading to economic losses and increased hazards to human and environmental health. Vegetables tend to have high insecticidal inputs because of their diverse insect pest complexes and high cosmetic standards. However, because of environmental and human health concerns, some older, broad spectrum insecticide classes are no longer being registered and this presents new challenges for insect pest management. To cope with these challenges there have been new advances in monitoring methods for detecting resistance and development of new insecticides with novel target sites. However, there has been limited success in implementing insecticide resistance management programs in vegetables. The reasons for this problem and potential strategies to overcome it will be discussed.

Transgenic crops for managing insect pests on vegetables: prospects and problems

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The area grown to transgenic crops for insect control was 18 million ha worldwide in 2003. However, only a small proportion of this total was grown to a vegetable crop- sweet corn. In contrast to field crops, development, commercialization, and deployment of biotech vegetable crops has taken dramatically different trajectories, including sharp declines in acreage, differential trajectories for fresh- and processing markets, and successful deployment of cultivars aimed at insect-transmitted pathogens. Biotech vegetables will present new challenges and opportunities for those involved in crop protection, marketing and consumption of insect- and insect-vectored pathogen protected plants. Much has already been learned about food safety, environmental and regulatory issues, and influence of adoption on pesticide use patterns from work with Bt corn, cotton, and potato, and from biotech papaya and squash. This may help pave the way for the development and deployment of biotech insect- or insect-vector pathogen - protected vegetables.

Cultural management of cotton aphids

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Studies were conducted in the Texas Rolling Plains from 1988 to 1994 to determine the influence of planting date on cotton aphid management. Dryland cotton was planted in late April, late May, and late June, and aphid numbers were consistently highest during August in cotton that was planted in late June. Percentage leaf nitrogen was highest during August in cotton planted in late June, but yields were lowest in cotton planted in late June. Nitrogen fertility rates should be adjusted in each planting date to minimize aphid numbers and to optimize yield. Aphid numbers were significantly reduced by dicrotophos applications during August in cotton planted in late April and late May, but control was inefficient in cotton planted in late June. Efficacy was influenced by leaf nitrogen and temperature conditions. Studies were conducted from 1997 to 2000 to evaluate date of irrigation termination on aphid populations during boll opening in September. Aphid numbers were lower in cotton that received a final irrigation in mid-August compared with numbers in cotton that received a final irrigation in late August. Aphids were more difficult to control with an insecticide in cotton that was irrigated in late August.

Studies on the flatid *Jamella australiae* Kirkaldy causing dieback in *Pandanus tectorius* var. *pedunculatus* (A.Br.) Domin on the Sunshine and Gold Coasts in southeast Queensland

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The flatid, *Jamella australiae* Kirkaldy, has caused extensive dieback in pandanus, *Pandanus tectorius* var. *pedunculatus,* on the Queensland Sunshine and Gold Coasts. *J.australiae* is a native species that has spread to these areas from north Queensland within the last 10-15 years. Flatid populations in North Queensland are low and non-damaging. A survey of 3200 pandanus in Noosa National Park during 1995 showed a high correlation between tree health and flatid numbers ($P<0.001$). The percentage of dead or poorly growing trees increased from 45.8% to 66.4% to 75% from 1995- 1997. By March 1998, 45% of trees had died. Predatory syrphids and lacewings, and an egg parasitoid, *Aphanomerus* sp.were ineffective. At Dimbulah in north Queensland, two other species, *Aphanomerus* sp.nr *pusillus* Perkins, and *Oencyrtus sp.,*were recorded parasitising up to 80% of the egg rafts and 80% of eggs within a raft. The Dimbulah parasitoids were released in southeast Queensland in 1996-97. *Ooencyrtus* sp established but wasn't effective. *A.* sp. nr. *pusillus* established at 20 release sites from where it dispersed throughout the Sunshine and Gold Coasts. In most instances, parasitism is at similar high levels to those at Dimbulah, and there has been an 80% reduction in flatid numbers.

Grower adoption of integrated pest management systems in tree crops in northern Australia

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There are few reports in the world-wide literature on the grower adoption of Integrated Pest Management (IPM) principles in tropical and subtropical tree crops. An email survey was conducted to determine the level of grower adoption of IPM in the most economically important tree crops in six regions across northern Australia. Recipients were experienced agricultural practitioners both in the private and government sectors and involved in the fields of plant health or pest management. In descending order, the six most important crops were mango, banana, citrus, avocado, macadamia and rambutan. The mean IPM adoption level (rated Low, Medium, High; 1-3 respectively) for the crops was 1.98. The highest adoption level of 2.67 was reported in rambutan while mango had the lowest with 1.53. Respondents considered that the IPM components most adopted by growers were 'soft' pesticides, regular monitoring, and plant health/nutrition in that order. The most important reason for adopting IPM was the growers' concerns about over-use of pesticides. Respondents indicated that the growers' main source of IPM information was from government departments and other growers. Various strategies that may assist in increasing grower adoption of IPM principles in these and other tree crops in northern Australia will be discussed.

Advances in managing Russian wheat aphid (*Diuraphis noxia* Mordvilko)

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The Russian wheat aphid (RWA), *Diuraphis noxia* (Mordvilko), has become one of the most damaging pests of wheat and barley globally, since its discovery in Russia in 1900. Characteristic RWA damage symptoms include leaf rolling, leaf folding, intervenal chlorosis and grain panicle trapping in the leaf sheath. Systemic insecticides have provided acceptable RWA control, but contact insecticides are ineffective against RWA colonies in rolled leaves. Cultural practices for RWA control include eliminating over-summering reservoirs, delayed planting in the fall, and early spring planting. Several RWA predators and parasites are abundant in the northwest U. S. and South Africa, and appear to have substantially reduced the incidence of RWA damage. Diverse complexes of entomopathic fungi lower RWA incidence on irrigated wheat in the U. S. (Colorado) and South Africa but not in dryland wheat. Resistant wheat cultivars have proven useful in managing RWA on wheat in Colorado and in South Africa. However, RWA field populations in several countries have developed virulence to different resistance genes, necessitating the identification of sources wheat resistance to the virulent populations. Data from successful RWA - IPM programs in different countries will be reviewed and discussed.

Molecular mapping of cereal aphid resistance genes in wheat

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Segregating wheat populations with different genes for resistance to greenbug *Schizaphis graminum* Rondani, biotype I or Russian wheat aphid *Diuraphis noxia* Mordvilko, (RWA) biotype A have been evaluated to identify molecular markers linked to resistance genes. The *Gbx, Gby* and *Gbz* genes express resistance to greenbug and are linked to molecular markers that provide selection accuracy for these genes ranging from 93.2% to 100%. Cytogenetic and physical maps indicate that *Gby* is located on the short arm of wheat chromosome 7A, while *Gbx* and *Gbz* are part of a resistance gene family located on the long arm of wheat chromosome 7D. In contrast, RWA resistance genes occur on the short arm of wheat chromosomes 7D or chromosome 1D. A gene family consisting of the allelic or tightly linked genes *Dn1*, *Dn2*, *Dn5*, *Dn6*, *Dnx,* and four uncharacterized *Dn* genes, is tightly linked to microsatellite marker *Xgwm111* on wheat 7DS. *Dn4* and an uncharacterized *Dn* gene, linked to *Xgwm106* and *Xgwm337* on chromosome 1DS, are either allelic or linked. A marker assisted selection system to identify aphid resistance genes in germplasm as well as in resistant plants under RWA attack is currently being developed.

Solanum viarum **Dunal. – a potential trap crop against tomato fruitworm (TFW),** *Helicoverpa armigera* **Hubner (Lepidoptera : Noctuidae)**

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Solanum viarum, a wild solanaceous plant was heavily infested by tomato fruitworm, *Helicoverpa armigera,* which opened the possibility of using this plant as a trap crop. Host plant selection in TFW is primarily a function of ovipositing female as newly emerged larvae are limited in their dispersal abilities. Hence, we aimed to explore the ovipositional cues of this plant and n-alkanes were identified as a major group. Though *S. viarum* was highly attractive to ovipositing females and young larvae, the later instars failed to develop, which ultimately resulted in death. In our experiments, the larvae did not feed on pure rice diet (check) and 60% of them died within a week. In diet fortified with *S. viarum* aqueous extract, only 20% could feed and survive up to four weeks. The larval duration was 25.60 days in diet fortified with hexane fraction of steam distillate (SD). The percent pupation (70.00) and pupal weight (0.139g) were significantly different in diet fortified with residual water fraction of SD. The adult emergence was only 20% where the larvae were fed with *S. viarum* extracts. The adults were unable to emerge completely. Hence, *S. viarum* can be used as a potential trap crop for managing TFW.

Impacts of insects on forest landscapes: implications for forest health management

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Change in forest landscapes occurs as a result the activities of humans and through natural disturbances. As human activities and intervention increase in frequency and impact, the consequences may be visualized in terms of deforestation and reforestation, corridors such as roads and powerlines, conversion of forested lands for agriculture, urban and suburban development, recreation and/or preservation, plus effects of atmospheric pollution and climate change. Humans may also modify natural rhythms of forest succession by lengthening normal cycles of fire or harvesting and replanting forests to select for individual species or age class structure. The resulting forest conditions affect population dynamics of native and introduced insects and may stimulate insect epidemics of amatic proportions. Although primarily aimed at reducing risk of fire, recently enacted legislation by the United States Congress, the Healthy Forests Restoration Act of 2003, is also aimed at ameliorating forest conditions that favor wide-scale insect epidemics. Examples of such outbreaks from North American forests currently abound. We discuss their causes, and examine the impacts of insects on resources and conditions of managed forest landscapes through examples that include red oak borer and southern pine beetle.

Selective control of chironomids and the preservation of biodiversity in Australian rice crops.

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The root-feeding larvae of chironomid midges cause major problems for Australian rice farmers. Chemical control is used in aerial sown crops on a routine basis during plant establishment, and often no other insecticides are applied to the crop for the remainder of the season. Chironomids are rarely a problem in drill sown crops, which are only intermittently flooded during crop establishment. Field studies have shown that aerial sown crops treated for chironomid control support lower levels of aquatic invertebrate biodiversity than drill sown crops. Despite major reductions in chemical use for chironomid control (> 90% in the last decade), these chemicals continue to be a threat to aquatic biodiversity. Recent studies show that *Bacillus thuringiensis israelensis* (BTI) has good potential for the control of *Chironomus tepperi*, which is known to be the principal pest species in rice. BTI shows a high degree of selectivity between different chironomid groups, and its potential use requires that we develop a better understanding of the feeding habits of other chironomid taxa (particularly *Polypedilum*, *Cryptochironomus* and *Tanytarsus*) present in the rice field environment. We are currently developing molecular techniques for investigating food preferences in chironomid larvae.

Hemipteran pests and their management in the US cotton belt

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Several hemipteran pests including *Lygus* species, clouded plant bug, cotton fleahoppers, and stink bugs are significant pests of cotton grown in the US Cotton Belt. These pests have increasing importance owing to a reduction in use of broad spectrum insecticides. This reduction is the result of boll weevil eradication efforts, the use of Bt technology, and greater reliance on relatively selective insecticides. This paper will review the basic biology of "bug" pests, with special emphasis on the mid-southern and southeastern US cotton production regions. The injury caused by these pests and experimental evaluation of various management approaches (such as insecticide applications, area-wide destruction of alternate hosts, and the use of trap crops) will be addressed.

The use of plant resistance for management of the rice water weevil in Louisiana

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The rice water weevil, *Lissorhoptrus oryzophilus* Kuschel, is the most destructive insect pest of rice in the United States, and has recently become a global threat to rice production through its introduction into major rice-producing regions of Asia. Current research on the use of host-plant resistance against this pest focuses not only on genotypic-specific resistance but also on the increases in resistance that occur as rice plants develop, irrespective of genotype. Variation in the tolerance of rice genotypes to feeding by weevils has been demonstrated in commonly-grown commercial cultivars and in a number of additional rice genotypes. Similarly, variation in the resistance of rice genotypes to infestation by weevils has been demonstrated in commercial cultivars and other rice genotypes. A breeding program for weevil resistance has been initiated. Furthermore, plant age-dependent increases in both the tolerance and resistance to infestation of rice plants have been shown. The efficacy of cultural strategies such as delayed flooding and early planting may be explained, in part, by these age-related increases in rice plant resistance.

Biology and management of *Batocera rufomaculata* De Geer (Coleoptera: Cerambycidae) in Jammu region of J&K State, India.

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The longicorn a long-horned beetle *Batocera rufomaculata* has been recorded as serious pest of large number of horticultural and forest trees. The author has made detailed observations on the biology and management of this pest on mulberry plantation. The salient observations revealed that adult beetles start emerging from the pupal chamber after the first monsoon shower and their number increases as the monsoon progresses. Adults feed by gnawing the bark or the green tips of the mulberry branches. During this period the male and female copulates and start deposition after carefully selecting suitable sites. Before deposition of an egg the gravid female makes a girdle around the selected twig, beyond which normally the branch falls with the action of the wind or it may subsequently y. Incubation period varies between 6-12 days. Newly hatched grub starts tunneling the twig/plant in a distinct pattern. In the process a large amount of frass is thrown out through a series of ejection holes, the number of which increases as the larva goes deeper in the branch. These ejection holes also serve as aeration holes. As the larvae grows, type of the frass changes. Distinct period of rest and the number of larval instars in two years life cycle of the beetle have been recorded. Based on the biological studies and screening of various chemicals, insecticides, different combinations and their effectiveness for the control of this pest at larval stage have been suggested.

Environmental risk associated with plant resistance to insects

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Insect-resistant crops represent a growing sector of the global agricultural scene, delivering economic and environmental benefits to growers and communities in diverse geographies around the world. Environmental benefits of Bt crops, such as reduced use of conventional broad-spectrum insecticides, can lead to more sustainable farming practices. These environmental benefits cannot be realised, however, until the technologies complete extensive regulatory review. Monsanto Australia achieved regulatory approval of Bollgard II® cotton in 2003, after years of research, development and regulatory activities in the US and Australia. This achievement, while significant, represents one milestone in the entire product stewardship lifecycle applied by Monsanto for its biotech products. Environmental risk assessment and management processes feature throughout the Bollgard II stewardship lifecycle: from early regulatory studies in the laboratory and glasshouse that determined safety to non-target organisms, to environmental impact studies in the field, and post-marketing stewardship such as the effective implementation of the Bollgard II resistance management plan. Moreover, after two commercial seasons of Bollgard II, the benefits of the technology to the Australian environment have also been demonstrated, with significant reductions in conventional pesticide use, leading to increased numbers and diversity of insects within the biotech crop.

Optional strategies for management of the sucking bug complex in US Cotton

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Historically, insecticide use has been heavy throughout the southern cotton belt. Following the USDA Boll Weevil eradication program and the introduction of B.t. cotton varieties, insecticide use decreased dramatically. New,"low-insecticide-input" cotton production systems are threatened by the rising importance of a complex of sucking bugs (e.g., tarnished plant bugs and southern green, brown, and green stink bugs), all of which were previously controlled by insecticides applied for boll weevil and lepidopteran pests. Growers report frustration that despite rising expenditures for bug control, yield and quality continue to decline because of bug damage. Current methods used to assess and quantify bug densities and associated damage are ineffective; thus, insecticide use seldom matches the actual need. Farmers are inclined to either over-treat with insecticides to protect their investment in B.t. cotton or revert back to non-B.t. production. Discussion will include bugs involved, their distribution, the need for dynamic treatment thresholds, and how bug and boll sampling can be simplified through application of technology. We will also examine the feasibility of site-specific applications to control bugs in cotton.

GM insect-resistance: pest management or mis-management?

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Host plant resistance (HPR) can be used as the central pivot of true IPM. It can synergise with biocontrol by a variety of routes, including the exploitation of the inherent selectivity of reduced-dose pesticide applications. The kinds of interactions between HPR, biocontrol and chemical control (the 'Pest Management Triad') can prevent or delay the development of pest tolerance to any of the three component control methods However, these valuable synergisms depend on the resistance being partial, and not based on allelochemicals, particularly single ones that are highly toxic. HPR based on high expression of a single toxin, the principle of current GM insect-resistance, has all the characteristics of pest mis-management inherent in the early insecticides such as DDT. Yet GM insect-resistance, although comparing badly with partial and broadly-based HPR, nevertheless compares rather favourably with insecticides, perhaps the fairest comparison. Surely GM is more often alternative technology to pesticides than to HPR?

Pesticide-free vegetables: commercially successful biological control in greenhouses

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Development of pesticide resistance in greenhouses initiated biocontrol research. The first natural enemies became available in the 1960s. A change from pure chemical control to advanced Integrated Pest Management took place in 20 years. Nowadays, growers annually introduce billions of natural enemies in greenhouses, and 125 species are commercially available for control of all important insect and mite pests. In the main vegetable crops in northern Europe, most if not all insect problems can be solved without the use of insecticides. The change to biocontrol was rooted on clear advantages for the grower. Integrated management of pests and diseases without conventional chemical pesticides is a goal that will be realized in Europe within a decade. Plant-breeding activities contribute to these IPM programmes by (1) combining host-plant resistance with biological control, (2) selecting plant cultivars that stronger attract natural enemies, and (3) selecting plant cultivars that produce a better "working environment" for biological control agents. Recently, the development of so called "closed system" greenhouses with strongly optimized energy management and nutrient recycling, resulted in improved biological control of pests and diseases due to a more stable greenhouse climate. (van Lenteren 2000 A greenhouse without pesticides. Crop Protection 19:375-384)

Semiochemicals – their role in the management of tropical tree fruit pests

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Semiochemicals are now in common use as pest management tools. They have application as a means of monitoring fluctuations in pest populations and so assist in the timing of control measures, and as control measures in their own right, in which they are released as mating disruptants or as attractants in 'attract and kill' formulations. But have they lived up to expectations? Interpretation of trap catch is notoriously difficult and mating disruption is not suited to all pests and situations. The impact of attract and kill formulations that attract only males may be limited, whereas those that attract females should, in theory at least, have better prospects of success. The various techniques are reviewed in the context of managing tropical tree fruit pests.

Cotton IPM in South America – progress and challenges

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IPM strategy to manage pests of cotton is broadly accepted and practiced throughout South America. The main cotton producing countries in the region are Brazil, Argentina, Paraguay, Bolivia, Perú, and Colombia. Few pests are common to most of the countries (e.g., cotton leaf feeder, *Alabama argillacea*, and pink bollworm, *Pectinophora gossypiella*). Other insects are restricted to some areas of few countries. Some countries have developed erradication programs against boll weevil, *Anthonomus grandis*. Lepidoptera resistant transgenic cotton is commercially available in Argentina and Colombia. Details on its efficacy, and level of acceptance from farmers will be presented, along with the IRM program in each of these South American countries. The progress made in the areas of pest management decision-making, evaluation of economic thresholds, and research on new chemicals, cultural control, biological agents, and biotechnology will be discussed with case studies from major cotton areas of South America. Information presented will be obtained from published papers, personal communications with researchers and extension specialists, and personal experience. Cotton arthropod pest complex, pesticide resistance issues, and specific pest management practices for different sizes of farms in each of these countries will be discussed, with special emphasis in Argentina situations.

Physical control methods for managing vegetable insect pests

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Physical control may offer innovative alternative to insecticides. After a brief review of the theory of physical control methods, we will discuss case studies from Canada (Quebec, New Brunswick and British Columbia) and Israel. Eastern Canada has played a significant role in the development of new physical control methods against agricultural insect pests since the mid 80s. Successful designs of plastic lined trenches, vacuums, propane flamers and others to control insect pests of the potato crop were achieved by combining entomological and engineering research interests. Reliance on chemical control methods has slowed down the adoption of physical control methods in traditional agriculture but the increasing place taken by organic production is creating opportunities. Israel has long been at the forefront of research on the use of physical control methods for agricultural pests. Major developments include: insect exclusion screens in the 1970s, which are now found covering greenhouses world-wide; colored plastic mulches, utilized since the early 1980s; and, recently, ultraviolet absorbing plastics, shown to be effective against several pests. Research is on-going in these and other areas. Future directions for research will be discussed.

Manipulating the phenotypic expression of resistance in young conifers

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Seedling conifers must grow fast enough to establish successfully in competition with other plants yet allocate adequate resource to defence against pests and pathogens. Environmental conditions that affect the growth of trees can alter the absolute and relative concentration of nutrients and carbon-based secondary chemicals and can also influence the physical characteristics of different parts of the tree. It is however, difficult to generalise about environmental influences on the phenotypic expression of resistance because the significance of these changes for different pests and pathogens will depend on their particular adaptations. The manipulation of plant growth to produce a range of phenotypes can, however, be a useful tool in the experimental investigation of 'quantitative' resistance traits and in determining the extent to which resistance expression could be manipulated in pest management. In this paper, the physical and physiological limitations on the phenotypic expression of resistance in seedling pine and spruce are discussed in relation to defence against the stem-feeding weevil, *Hylobius abietis*. Results are presented of the effect of light and nitrogen treatments of young trees on the resin duct system and consequences for feeding and reproductive behaviour of adult weevils.

Exclusion nets - a viable management option for vertebrate and invertebrate pests of tree crops

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Vertebrate pests have become a chronic problem for the expanding areas of mono-cultured tree fruit crops, especially in tropical and subtropical Australia. Birds and bats have multiplied under environmental protection legislation in Australia, despite a reduction in some natural habitats. Protected populations require more food than native sources can provide to sustain increasing numbers, and fruit crops provide a bountiful source that is easily accessed. Various counter measures have been used to mitigate vertebrate pest damage. Flashing lights, noise, high voltage electrical grids, and shooting, have all been tried. Some of these are now illegal, and increasing urbanisation limits the use of most. Exclusion netting is the only viable option remaining, and is being used successfully in commercial orchards. The development of new technology in net manufacture and erection has initiated interest among entomologists with respect to the potential exclusion of many insect pests. Some pests are excluded by nets used to protect crops from hail, and total exclusion of fruit flies from stone fruit has been achieved in Queensland trials, using finely woven nets. In addition to achieving effective pest exclusion, the physiological effects on the crop of shading, reduced wind movement and elevated temperatures must also be considered.

Improving vegetable IPM with introduced parasitoids: successes and challenges

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IPM programmes for process tomatoes and vegetable brassicas in New Zealand are based on cost-effective monitoring systems, validated action thresholds and strategic use of selective insecticides to maximise the impact of introduced larval parasitoids. *Cotesia kazak,* the dominant parasitoid of tomato fruitworm in tomatoes, is well synchronised with its host and kills larvae while still small, preventing major fruit damage. Other introduced parasitoids complement *C. kazak*, and when combined with use of an adjustable action threshold using estimated larval parasitism, have led to a 90% reduction in insecticide use. In vegetable brassicas the recently introduced parasitoid of white butterfly, *Cotesia rubecula*, is displacing the earlier introduced *Cotesia glomerata*. *C. rubecula* is the superior competitor and better synchronised with host populations. Along with *Diadegma semiclausum* attacking diamondback moth, these two parasitoids can attain high levels of parasitism. In combination with well-timed larvicides, insecticide applications may be reduced by 75%. Challenges include selecting effective parasitoids, minimising non-target effects, integrating parasitoids with insecticide use, and maintaining adequate support systems for the IPM programmes.

Apparent genotypic resistance in Norway spruce to feeding by the weevil *Hydrobius abietis*.

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The large pine weevil *Hylobius abietis* L. is a major pest in conifer plantations in the palearctic region. Field studies in Sweden on 52 clones of Norway spruce *Picea abies* L. (Karst.) indicate that there is a genetic factor involved in tree resistance/tolerance. At present growth parameters are used for genotype selection in the Swedish spruce breeding program. The results suggest that it is important to study trade-offs between these growth parameters and resistance to pine weevils

Recent stem borer research on rice in the United States

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Stem borers, primarily sugarcane borer (*Diatraea saccharalis*) and Mexican rice borer (*Eoreuma loftini*), are becoming increasingly problematic in the southern United States. Recent investigations in Texas have shown that *E. loftini*, introduced from Mexico into Texas in 1980, is spreading north and east towards Louisiana where production of rice and sugarcane is threatened by this exotic pest. Cooperative research between Texas A&M University and Louisiana State University is aimed at developing a pro-active IPM program for stem borers attacking both rice and sugarcane. The program includes studies on host plant resistance and cultural and chemical control of stem borers. In regards to rice, results of studies have demonstrated that 1) Cocoie, the most commonly grown rice variety in the southern United States, is very susceptible while hybrid rice cultivars are least susceptible to stem borers; 2) stem borers do not attack rice before panicle differentiation; and 3) transgenic rice shows great promise as a genetic method of control. In addition, research results have led to the registration of lambda-cyhalothrin for stem borer control. Recommended rate is 0.034 kg (AI)/ha applied twice at approximately panicle differentiation and late boot.

Compensation for pest damage – linking pest and damage based thresholds

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Cotton compensates for pest damage, often with little effect on yield or maturity. Damage thresholds, if developed, combined with pest thresholds, would account for both plant recovery and potential pest damage thereby avoiding control of non-economic pest densities or damage. Experiments using leaf removal to simulate pest damage found young cotton plants could tolerate >80% early leaf loss without affecting yield or maturity. Similarly, manual removal of apical meristems from 100% of plants three times or of the apical meristem plus two nodes from 100% of plants did not effect yield and delayed maturity <5d. Flower bud removal from the first 4 fruiting branches did not affect yield but caused a delay of 7d. Effects of leaf and terminal damage on yield and maturity were additive and reasonably predicted by multiple regression equations. These have been used to develop tentative damage thresholds. Distribution of damage between plants was less important than amount of damage in determining recovery. When leaf damage was inflicted on 0%, 25%, 50%, 75% or 100% of plants the performance of undamaged plants changed, allowing compensation. Modelling compensation has begun using the CSIRO OZCOT cotton crop simulation model to provide greater flexibility for exploring and predicting compensation.

Southern hemisphere exotic pine plantations threatened by insect pests and their associated fungal pathogens

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Plantations of exotic *Pinus* spp. in the Southern Hemisphere cover many millions of hectares and represent a major industrial resource. The success of these plantations is commonly attributed to the fact that the trees have been separated from their natural enemies, including insect pests and pathogens. Recent trends show clearly that introductions of new pine pests and pathogens are increasing. This is a situation that seems likely to continue as the movement of people and products increases. Trends also suggest that once an insect pest or pathogen has become established in a new Southern Hemisphere environment, the likelihood of movement to adjacent countries and continents increases. Here, patterns of movement of the Sirex woodwasp *Sirex noctilio* provide an interesting example. Interactions between fungal pathogens and introduced insect pests are also appearing. For example, the damaging pitch canker pathogen *Fusarium circinatum* has been introduced into nurseries in various southern hemisphere countries. Associations between this pathogen and various insect pests are emerging and these are likely to heighten an already serious impediment to pine propagation. Future forest management practices will need to incorporate a clear understanding of these threats, if profitable exotic pine propagation is to be sustained in the long term.

Long-term benefits of the integrated management of rice insect pests in Dasha Town of Sihui City, Guangdong Province, China.

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A biocontrol-based IPM program has been conducted since 1973 in the rice ecosystem in Dasha Town of Sihui City of Guangdong Province, China. Experimental areas of the program were extended from 1.6 ha in 1973 to about 4000 ha in 1975. Experts from the university passed on IPM knowledge and measures to farmers through Farmer Field School (FFS) in intervals. IPM measures have since then been adopted conscientiously by the farmers. The program resulted in significantly ecological, social and economic benefits, including (1) the population of major insect pests in the rice field obviously decreased; natural enemies had successfully restored and kept the dominant position, which play a key role in the control of rice insect pests; (2) the amount of pesticides used in the rice field was decreased dramatically; and the pesticide residue of the rice and soil in Dasha, measured in 1983, 1993 and 1997, has showed very low and met the State Criteria for "Green Food". As a result, the rice product in Dasha Town was allowed in 1998 to be labeled "Green Food" (LB-18-9801190913).

Paradigm shift in pest management in rice in southeastern China newly invaded by the rice water weevil *Lissorhoptrus oryzophilus* Kuschel (Coleoptera: Curculionidae)

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The alien rice water weevil (RWW) has been severely threatened to the rice production in the SE coaster Zhejiang, China. There was only one generation in one season of rice. Most of the adults emerged from 1st-season rice was in reproductive diapause and migrated to hills for oversummering and overwintering, while only minors migrated into newly grown rice of 2nd-season rice and developed 2nd-generation. Species and age of host plant, and adult density may change the proportion of reproductive females. Compared with the management measures for insect pests before RWW invasion, nowadays two extra insecticide applications are implemented by farmers: control of the overwintered adult weevils by triazophos, fipronil or fenvalerate in late April to early May in seedling beds or early transplanted fields, and control of 1st-generation adults by triazophos, trebon in late June to early July. The larval populations of 1st-generation RWW and stem borers can be controlled by triazophos in mid - late May simultaneously. Because of the risk of resurgence of planthoppers resulted from triazophos and pyrathroids via killing of natural enemies and stimulation of reproduction, conflict between loss of ecological function and efficient of control of RWW must be solved in a balanced way.

Integrated Pest Management of Hessian fly (*Mayetiola destructor* Say)

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Hessian fly, *Mayetiola destructor* (Say), is the most destructive insect of wheat throughout the world. It is an economical pest in Morocco, Tunisia, Algeria, Kazakhstan, Spain and the USA. When heavy infestations occur on young tillers, damage can result in total yield loss. In Morocco, losses were estimated to 36% and 32% of bread and durum wheat yields, respectively. Integrated pest management uses a variety of methods to keep crop damage below an economic threshold without causing adverse effects on the environment. It reduces the selection pressure and enhances the durability of resistance. Several methods of control are being integrated to manage hessian fly. Host plant resistance is the most practical and economic method of control. Sources of resistance are continuously sought and incorporated into high yielding wheat cultivars. Thirty one genes of resistance have been identified in the usa, out of which 10 (*h5*, *h11*, *h13*, *h14h15*, *h21*, *h22*, *h23*, *h25* and *h26*) are effective in morocco, two (*h25* and *h26)* and three (*h13*, *h25* and *h26*) confer resistance to the syrian and the kazakh hessian fly populations, respectively. The genetic interaction between wheat and hessian fly is highly specific and consistent with a gene-for-gene relationship. Therefore, biotypes evolve, like it has happened in the usa. Other methods of control such as chemical control, rotation, planting dates, and nitrogen top dressing are also practiced to reduce the fly damage.

Laboratory rearing techniques for the giant wood moth (*Endoxyla cinerea*) and a longicorn beetle (*Phoracantha solida*) in Queensland.

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In eucalypt plantations in southeast Queensland a number of key insect pests have been identified causing damage. Among the most important of these are two stem boring insects, giant wood moth (*Endoxyla cinerea*) and longicorn beetle (*Phoracantha solida*). Basic biological information on the lifecycles of these insects is limited but was required to support research into identifying effective management methods. Cultures of both these species were established in 2001. Wood moth larvae were initially sourced as relatively mature larvae extracted from infested trees and reared on an artificial diet based on wheatgerm blended with host bark. Later, first instar larvae reared from eggs were also established on the same diet. Generally, late instars did not accept the diet well and suffered considerable fungal mortality. Early instar larvae accepted the diet but there was limited success in rearing them through to later instars. However for both groups the process was labour intensive and was not an economically viable method for obtaining adults for experimentation. In contrast, longicorn larvae reared from eggs accepted the diet well, had fast growth rates, low later instar mortality, and required less frequent handling. *P. solida* have now been reared for three generations in the laboratory.

Managing leaf beetles in eucalypt plantations: non-target impacts, biocontrol and natural enemy recolonisation

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A field trial examined the impact of chemical spraying on leaf beetle (*Chrysophtharta agricola*: Chrysomelidae) control by natural enemies in *Eucalyptus nitens* plantations. We compared the impact of the Naturalyte insecticide Success$^{\text{Ò}}$ with the broad-spectrum pyrethroid Dominex$^{\text{Ò}}$. In addition to reducing natural enemy loss to insecticides we hypothesised that we would enhance natural enemy effectiveness and residency in the plantation. Non-target impacts were monitored using op sheets during aerial spraying, sticky traps, malaise traps and visual searches of branches before and at intervals after spraying. The efficacy of natural enemies was assessed by protecting sentinel cohorts of beetle larvae during spraying and monitoring their disappearance due to predation or levels of parasitism. As anticipated, Dominex$^{\text{Ò}}$ reduced levels of non-target species, including natural enemies, more than Success$^{\text{Ò}}$. However, predation rates were higher following Dominex$^{\text{Ò}}$ spraying with the few recolonising and remaining resident predators targeting the sentinel larvae. By contrast, predation rates following Success$^{\text{Ò}}$ spraying mirrored that of unsprayed plantations. Parasitism rates of sentinel larvae by tachinid flies were severely impacted by either spray, though less so under Success$^{\text{Ò}}$. The return to pre-spray levels of natural enemies varied according to species but was often at least two months following broad-spectrum spraying.

Investigations into the effect of four insecticides on taro beetle (*Papuana Uninodis* Prell) infestation on taro (*Colocasia Esculenta*) in two selected sites in Fiji

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Taro beetle (*Papuana uninodis* Prell) was first recorded near Suva in 1984. *P.uninodis* creates tunnels on taro corms and severely reduces its quality. The beetle has spread to taro growing areas in most parts of the Southern side of Vitilevu, Ovalau Island and was recently found in Vanualevu, poses a very serious threat to taro production in Fiji. Field trials to determine the effect of Suscon Indigo, Confidor and Chlorpyrifos insecticides were conducted in taro beetle infested sites in Ovalau and Navua in 2002-2003. In addition, Nutri-gro-plus insecticide was tested only in Navua. Each insecticide treatment was replicated five and six times in Ovalau and Navua respectively and the trials were laid out using the Randomised Complete Block Design. The highest total corm weights from both field trials were observed in the Confidor-treated plants, which out yielded ($P<0.05$) the Chlorpyrifos-treated and Suscon Indigo-treated plants and Nutri-gro-plus in Navua only. In both field trials, the Suscon Indigo insecticide did not perform well in minimizing the damage caused by the beetle as compared with the untreated-plants at ($P<0.05$). Therefore, treating taro plants with the recommended rate of Confidor insecticide resulted in higher market corm weights.

Adapting and validating spatially variable insect control programs for cotton

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Geographic information system data are being correlated with in-field densities of cotton insect pests and evaluated as a basis for variable rate pest control strategies. Insect pest densities are being correlated with remotely sensed images of variable plant biomass (Normalized Difference Vegetative Index or NDVI) and NDVI based spatially variable insect control programs evaluated. Insect pest populations that have reached sufficient density for evaluation include the cotton aphid (Hemiptera: Aphididae), tarnished plant bug (Heteroptera: Miridae) and bollworm/tobacco budworm (Lepidoptera: Noctuidae) complex. Correlations between tarnished plant bug densities and NDVI appear very transient. That is, good correlations are observed on some dates while correlations on other dates correlations appear negative. No apparent correlations between cotton aphid or cotton bollworm/tobacco budworm complex densities have been observed on any sampling dates. Yield impact of NDVI based insect control programs is still being evaluated. Historical crop yields from yield monitors are also being evaluated as a method to base variable rate insect control programs. No significant impact on crop yield has been observed when spatially variable treatments are based on historical crop yield or itability. Historical crop yields appear to be viable basis for spatially variable cotton IPM.

Field observations of the pasture mealybug, *Balanococcus poae* **(Maskell) in New Zealand**

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The pasture mealybug, *Balanococcus poae* (Hemiptera: Pseudococcidae) was originally identified on common tussock grass, but is now known to be endemic on a wide range of pasture grasses in Canterbury, Nelson, Manawatu, and other regions in New Zealand. *B. poae* feeds in the crown and upper root zone where it withdraws sap from plant tissues causing a general browning of the paddock which is often misidentified as drought stress. Damage can first become apparent in January during dry periods, but is usually most severe in autumn and early winter. A preliminary report published in 1988 by W.D. Pearson suggested that *B. poae* was univoltine with parthenogenetically produced offspring. Recent field studies conducted at AgResearch in Lincoln, NZ, however, revealed that *B. poae* has at least two generations per year. First and second generation crawlers reached maximum numbers in mid-November 2003 and mid-January 2004, respectively. Male mealybugs were present beginning in mid-December 2003, and reached peak abundance on 31 December. DNA analysis confirmed that collected males were *B. poae*. Further research is needed to fully understand the biology and develop effective management strategies for this hidden threat to New Zealand pastures.

Thrips and their management in the Mississippi River Delta cotton region

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Several thrips species, including *Frankliniella* species, are annually significant pests of cotton grown in the Mississippi River Delta Cotton Region. These insects are important early-season pests that can slow plant growth, and thus increase the crop's exposure to later-season pests and an early fall frost. This paper will review the species composition of thrips complex in cotton, with a special emphasis on the US Mississippi River Delta cotton production region. The injury caused by these insects and experimental evaluation of the various insecticide management programs (i.e. seed, in-furrow, and foliar treatments) will be compared and discussed.

Plant incorporated Bt resistance in maize for control of *Spodoptera frugiperda* and *Helicoverpa zea* in the southeastern United States

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Fall armyworm, *Spodoptera frugiperda* (J. E. Smith), and corn earworm, *Helicoverpa zea* Boddie, perennially cause leaf and ear damage to maize in the southeastern United States. Transgenic Bt hybrids with the Bt11, MON810, or 176 events expressing the Cry1Ab insecticidal endotoxin from *Bacillus thuringiensis* (Bt) were evaluated for control fall armyworm and corn earworm on 2-3 planting dates at seven locations in Georgia during 1999 and 2000. All Bt events consistently reduced whorl infestation and damage. Hybrids containing Bt11 and MON810 events reduced ear infestations in all trials, although reductions were small in later plantings. Because of lack of toxin expression, the hybrid containing Event 176 did not reduce ear infestations and damage. Total grain aflatoxin concentrations were not significantly affected by Bt resistance in any trial (N=17). Yield responses were variable with the prevention of yield loss being proportional to the severity of insect damage. Although Bt resistance did not consistently benefit recommended time plantings, Bt11 and MON810 events were effective in preventing significant losses to field maize when damaging infestations occurred. The Bt11 and MON810 events mitigated the risk of severe lepidopteran damage to maize, thereby making later plantings of maize feasible in double-cropping systems.

Extrinsic resistance potential for biologically-based turfgrass pest management

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Interactions between host plant resistance and biological control may benefit or hinder pest management. Turfgrasses have rarely been tested for extrinsic resistance characteristics -occurrence and performance of beneficial arthropods on plant genotypes with resistance to known turf pests. Among six turfgrass genotypes tested, the braconid wasp *Aleiodes laphygmae* Viereck, varied in percent parasitism of fall armyworm, *Spodoptera frugiperda* (J. E. Smith), larvae A total of 20,400 first instar larvae were placed in the field; 2,368 were recovered; 468 parasitoids were subsequently reared; 92.2% were *A. laphygmae*. The consistent dominance of *A. laphygmae* in the turfgrass environment is a departure from the minor role observed for this species in agricultural crops such as corn and sorghum. Parasitism by *A. laphygmae* was consistently highest on paspalum followed by bermudagrass and then zoysiagrass. Percent parasitism of *S. frugiperda* in this field study decreased with increasing resistance levels to the host insect. However, substantial parasitism on TifSport, a cultivar of bermudagrass demonstrating intermediate levels of resistance to fall armyworm, suggests a promising potential synergy between modest levels of host resistance and pest suppression by natural enemies. Additional studies demonstrated that turf taxa also roundly influences predator occurrence and performance and armyworm susceptibility to insecticides.

Development and parasitism of *Aphidius colemani* and *Lysiphlebus testaceipes* (Hym.: Aphidiidae) on *Aphis gossypii* (Hem.: Aphididae)

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The parasitoids *Aphidius colemani* and *Lysiphlebus testaceipes* are the most frequent natural enemies of *Aphis gossypii* under greenhouse conditions. The temperature directly affects the development time and behavior of insects. The adaptability to climatic conditions is a key point for the success of mass rearing and parasitoids establishment in biological control programs. The objective this study was to evaluate the development and parasitism of *A. colemani* and *L. testaceipes* on *A. gossypii* as a host, in controlled climatic chambers. *L. testaceipes* development on *A. gossypii* at different temperatures (15, 20, 25 and 30ºC) showed the shortest development time (11.3 days) and the highest percentage of parasitism and emergence (> 60%) at 25ºC. The rates of mummies and the emergence of *A. colemani* and its longevity decreased with the increase of the temperature (16, 19, 22, 25, 28 and 31ºC), and no mummy's formation at 31ºC. The results indicate that the temperatures 22ºC and 25ºC, respectively, for *A. colemani* and *L. testaceipes*, could be the most adequate for reproduction and establishment of each parasitoid as a biological control agent of *A. gossypii* in protected cultivation.

Olive lace bug *Froggatia olivinia* Froggatt (Hemiptera: Tingidae) – an Australian native pest of cultivated olives

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The olive industry has been the fastest growing horticultural activity in Australia over the past decade, with more than 8 million trees now in the ground. The native phytophagous tingid, olive lace bug, *Froggattia olivinia* has moved from its original host *Notelaea longifolia* to become an important pest of cultivated olives, *Olea europea*. Infestations of nymphs and adults suck sap from undersides of leaves, often resulting in severe damage or the death of host plants. *F. olivinia* was originally reported by Froggatt, in the only published paper on this species, from coastal and tableland regions of the eastern mainland, but it has since spread to inland areas of the eastern states as well as to Victoria and South Australia. An incursion of *F. olivinia* was detected in Western Australia in 2002, where a successful eradication campaign was conducted. This poster paper will present descriptions and images of *F. olivinia* adults and nymphs together with their damage.

"Seasonal fluctuations in crop damage of hemipteran pests in California pistachio"

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California pistachios are attacked by a variety Hemiptera, most notably stink bugs such as the *Thyanta pallidovirens* and *Acrosternum hilare*, and a leaffooted bug (*Leptoglossus clypealis*). We clarified seasonal changes in nut damage using cages that isolated adult hemipterans on pistachio clusters for 7 day periods (fruit set – harvest). Results show early-season feeding does not result in crop loss because the pistachio compensates for opped nuts. For example, pistachio clusters naturally declines from 80 to 20 nuts during the season. There are critical mid- and late-season periods when damage (epicarp lesions) because the natural nut op has been completed and Hemiptera feeding impacts the cash crop. For example, in late-May and early-June trials, a single stink bug feeding for 7 days caused about 40 – 50% damage of the available nuts (opped nuts or epicarp lesion) of cages nuts. Trials conducted later in the season, June to August, show fewer opped nuts and epicarp lesion. Finally, after August, it was difficult to determine damage levels by examining nuts for epicarp lesions because feeding wounds did not form for many weeks or months while the internally-damaged nuts remained in the cluster. Results have been used to improve seasonal insecticide applications.

Effect of temperature on activity of insecticides to the differential grasshopper – (Orthoptera: Acrididae)

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We evaluated insecticides for activity to third instar *Melanoplus differentialis* (Thomas) at temperatures from 10° to 35°C. In the laboratory, treatment with esfenvalerate resulted in 100% mortality and activity was not temperature dependent. Treatment with spinosad resulted in similar mortality as esfenvalerate at all temperatures except 10°C. The activity of *B. bassiana* was greatest at 25°C, and was adversely affected by high and low temperatures. Treatment with diflubenzuron resulted in increased mortality at high temperatures. Activity of azadirachtin was not influenced by temperature. In field studies, spinosad, diflubenzuron, endosulfan and esfenvalerate provided significant levels of mortality both at-application and when nymphs were exposed to 1-h old residues. Spinosad and diflubenzuron provided similar levels of mortality when nymphs were exposed to 24-h old residues. Residual activity of endosulfan, naled, esfenvalerate, and spinosad decreased with increasing time (0 to 24 hours) after exposure to sunlight and high summer temperatures. Compared to other insecticides, naled had a short residual activity period and activity was dependent upon immediate contact with the nymphs. *Beauveria bassiana* was inactive under high temperatures and intense sunlight as occurs in summer.

Identification and characterization of peroxidases in resistant sorghums

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Greenbugs, *Schizaphis graminum*, are important insect pests of sorghums in the United States. The identification of sorghums with resistant traits to greenbugs has provided a sustainable, economical and environmental responsible approach for managing this serious pest. However, information regarding the mechanisms of the resistance remains unknown. Previous research investigated the role of several oxidative enzymes (catalase, polyphenol oxidase and peroxidase) in the defense response of sorghum to greenbug feeding. This research documented no change in the activity levels of catalase and polyphenol oxidase levels in control and greenbug infested sorghum plants. However, there was an increase in peroxidase activity in resistant sorghums being fed upon by greenbugs. These results suggest that increases in peroxidases may accompany insect feeding in resistant genotypes and could be an important marker for selecting resistant germplasm. Our current research focuses on purifying the peroxidases present in resistant sorghums and characterizing their role in the defense response of sorghum to greenbugs. Results from this research will be presented at the International Congress of Entomology.

SuccessÒ is an alternative to DominexÒ for managing leaf beetles in eucalypt plantations

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A field trial compared the efficacy of the Naturalyte insecticide SuccessÒ (a.i. spinosad) with the pyrethroid DominexÒ (a.i. cypermethrin) for managing the eucalyptus leaf beetles, *Chrysophtharta bimaculata* (Olivier) and *C. agricola* (Chapuis) (Coleoptera: Chrysomelidae), attacking *Eucalyptus nitens* plantations. Larval and adult populations were monitored before and at intervals after aerial spraying using tagged shoots, sticky traps and visual assessments. Defoliation levels were assessed at the end of the summer. Non-target species were also assessed (see Allen *et al.* Poster). Both DominexÒ and SuccessÒ reduced egg and larval populations of *C. agricola* immediately following spraying and reduced the level of defoliation by larvae during the summer months. In the field, effective control of young beetle larvae from the spray residue on the foliage lasted approximately one week longer for DominexÒ than for SuccessÒ. Both DominexÒ and SuccessÒ were equally effective against the 1st instar larvae, while SuccessÒ was less effective than DominexÒagainst the 2nd instar larvae. Leaf beetles recolonised and laid more eggs within one month after spraying with SuccessÒ and within two months after spraying with DominexÒ. For maximum efficacy, application of SuccessÒ should be timed immediately after egg hatch when the 1st instar larvae are most abundant.

Insect management in litchi orchards: a tool for sustainable land use

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In litchi growing systems in northern Thailand frequent use of pesticides is the common farmers practice to prevent negative impacts of pest arthropods. Considering the ecology of the major pests it seems doubtful that they can be effectively controlled by this measure. However, it is well-known that the massive use of broad-spectrum insecticides leads to shifts in the composition of the arthropod community, i.e. not only pests are reduced, but also beneficial organisms such as pollinators, predators and parasitoids. To establish a sustainable land use management and to increase in arthropod biodiversity, which can also help to suppress pest populations, a reduction of pesticide application and a suitable vegetation management are required. In the present study the spatial and temporal patterns of the major pest and beneficial species are analyzed on landscape level. In a litchi orchard four different treatments including pesticide application (yes/no) in combination with mowing (yes/no) of the underground vegetation are conducted. Effects of the different treatments on the presence and diversity of beneficial insects are studied. Detailed knowledge of the seasonal dynamics and the insect antagonist-complex are expected results. In addition an analysis of relationships between the diversity of beneficial insects and the underground vegetation will be conducted.

Development of a numerical simulation model for long-range migration of rice planthoppers

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Trans-oceanic migration of rice planthoppers was precisely simulated by incorporating some parameters related to the flight behavior with a high performance atmospheric dispersion model. The model consists of an atmospheric dynamic submodel for calculating meteorological fields and a particle random-walk submodel for atmospheric dispersion. The model also functions in specifying source term information such as the source area, take-off time and the relative amount of the planthoppers. We carried out a case study of source term estimation for the migration of planthoppers to western Japan with net-trap catches obtained at nine locations in mid-June 1998. In the simulation, 56 areas each with the width of two degrees of latitude and longitude were set as the take-off area. Take-off duration time was assumed as one hour at dawn and dusk. Relative immigrant density originated from take-off area was calculated for each observation site, and rank correlation coefficients were used for the model evaluation. Possible migration source areas were distributed around 23-27N, including Fujian and Taiwan. Air temperature that allows the planthopper to fly continuously, and flight duration are critical parameters for the simulation results.

Managing stink bugs in cotton in southeastern USA

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Because stink bugs continue to pose a challenge to current and future efforts concerning cotton insect management, we continued investigations into alternative monitoring strategies and management tactics for the pest complex. Pheromone trapping of stink bugs was useful in following in-field populations of stink bugs, but the reduced availability and considerable expense of currently available lures and unavailability of lures for other important species continues to make potential pheromone trapping prohibitive. Research with treatment thresholds for stink bugs, based on monitoring internal feeding injury to bolls, supported treatment at the 10-20% injury to mid-sized (ca. 14-d-old) bolls. In laboratory bioassays, methyl parathion and dicrotophos provided superior control (94-100% mortality) of field-collected fifth instars and adults of the green stink bug (GSB), *Acrosternum hilare* (Say) and the brown stink bug (BSB), *Euschistus servus* (Say), at 0.5 lb (AI)/A. Pyrethroid insecticides provided variable results (11-100% 24-hr mortality) due to considerable tolerance by BSB. Results from studies addressing simulated mechanical injury to bolls suggested that losses from bug feeding injury to small-to-medium-sized bolls could be significant under certain circumstances. In caging experiments, the ability of GSB to damage cotton bolls and reduce yield decreased as bolls aged, and yields from bolls that accumulated 583 HU at 27 d following anthesis were not significantly reduced.

Biology and behavior of western leaf-footed bug on satsuma orange in USA

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The western leaf-footed bug, *Leptoglossus zonatus* (Dallas) was first noticed attacking Satsuma oranges in Plaquemines Parish, Louisiana in autumn 1997 and since then has become a serious pest. This species typically begins feeding on Satsuma in mid-August and continues feeding through harvest. It causes concealed damage to Satsuma through transmission of the yeast, *Nematospora coryli* Peglion. Studies were conducted to find an optimum diet for rearing this insect in the laboratory. Results indicated that the combination of fresh raw peanuts, potatoes and Satsumas was the most suitable diet. When given a choice of green beans, potatoes, sweet potatoes, fresh raw peanuts, Satsumas, and carrots first instar nymphs preferred to feed on Satsumas. Second through fifth instars and adults fed frequently on fresh raw peanuts, Satsumas and potatoes. Field observations in October 2003 revealed that adults moved around the tree during the day in order to remain in full sunlight. Fifty six percent of adults under observation in the field did not move during the night from their afternoon location. In the laboratory, both sexes fed more during scotophase than photophase. Females fed longer than males but males moved between fruits more often during a 24 hr study period.

Bioefficacy of azadirachtin against *Scirtothrips dorsalis* **infesting tea crop**

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India is the world's largest producer of tea, having 507200 hectares of land under cultivation. At field, several pests nestled on the tea crop canopy, affecting the production and quality of the made tea. One among them is *Scirtorthrips dorsalis* infesting two leaves and a bud as well as three leaves and a bud crop.

The present study was conducted to know the bio efficacy of Azadiractin 0.15EC (NEEMGOLD) against *Scirtorthrips dorsalis* at Hope Tea Estate, the Nilgiris district, Tamilnadu. Azadirachtin 0.15EC (NEEMGOLD) which is environmentally safe formulation, have the ingredient Azadirachtin-A, which is a tetranortiterpenoid and most active limonoid with insect growth regulating and antifeedant properties. The spray of Azadiractin 0.15EC (NEEMGOLD) at the rate 1.25 lit. per hectare at the first sight of *Scirtorthrips dorsalis* was found most effective for its control. Whereas, minimum thrips incidence was recorded with continuous three spray round. Besides thrips control, zero phytotoxicity was also observed on tea crop canopy, during the spray schedule.

Management of *Scirpophaga nivella* F. (Lepidoptera: Pyralidae) in sugarcane (Punjab, Pakistan)

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Ten varieties of sugarcane were screened with CP-43/33 and L-118 as susceptible, BF-162 and SPSG-26 resis-tant while CP-72/2086 as intermediate, for management studies of *Scirpophaga nivella*. Morpho-physio chemi-cal plant factors as well as quality of sugar were determined to see their affects on the pest infestation. Leaf thickness and moisture contents showed positive/significant correlation with the pest infestation at tillering stage. Total mineral, manganese and copper contents did not show significant correlation with the pest infesta-tion, whereas nitrogen, potassium, calcium, magnesium and ferrous contents manifested positive and sig-nificant contribution with the pest infestation. Phosphorous, carbohydrates, fats and zinc produced significant and ad-verse effect on the pest infestation at tillering stage. Zinc contents with contrasting behaviors appeared to be the most important character with co-efficient value of 0.764 followed by ferrous with positive sign. The effect of borer infestation was significantly negative on POL, Brix contents CCS. The coefficient of determination value was 0.821, obtained by computing fiber content, POL, Brix and CCS factors together for multivariate regression models. Application of trash mulching at the time of sowing proved to be the most effective treatment followed by Furadon, removal of dead hearts+spike trust, hand collection of egg masses and cutting of shoots at 15 days interval.

Evaluating Bollgard 2, WideStrike, and VipCot against lepidopteran pests in mid-southern U.S. Cotton Fields

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Bollgard$^{\text{Ò}}$ cotton cultivars produce the Cry1Ac protein from *Bacillus thuringiensis* Berliner var. *kurstaki* (Bt), which exhibits toxicity to larvae of many lepidopteran cotton pests. The Bollgard$^{\text{Ò}}$ technology has been very effective against tobacco budworm, *Heliothis virescens* (F.). However, Bollgard demonstrates limited activity against other lepidopteran pests including bollworm, *Helicoverpa zea* (Boddie); armyworms, *Spodoptera* spp.; and soybean looper, *Pseudoplusia includens* (Walker). Monsanto's new Bt cotton (Bollgard 2) was derived by incorporating the Cry2Ab protein into Bollgard cotton cultivars. Transgenic cotton lines (WideStrike) from Dow AgroSciences express the Cry1Ac and Cry1F Bt proteins. Syngenta's lines (VipCot) express a Bt protein that is different in structure and mode of action than proteins in other transgenic cottons. The objectives of these studies were to document technology efficacy against a spectrum of caterpillar pests and to determine their role in a cotton IPM system. The Bollgard, Bollgard 2, WideStrike, and VipCot technologies demonstrated satisfactory control of tobacco budworm. Bollgard 2, WideStrike, and VipCot generally provided acceptable control of bollworm and other defoliating pests, including beet armyworm, *Spodoptera exigua* (Hubner), and soybean looper. These technologies will further reduce, and in some instances, eliminate a requirement of supplemental control of caterpillar pests with foliar applications of insecticides.

Characterization of bruchid resistance in the mungbean variety VC6089A and the resistance-associated protein VrD1

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A novel bruchid-resistant protein, VrD1, was identified in the mungbean variety VC6089A, which was bred from a wild accession TC1966. In the present study, the resistance of VC6089A variety and the toxic-related protein VrD1 was investigated against the cowpea weevil, *Callosobruchus maculatus*. We found that the seeds of VC6089A showed great resistance by causing more than 96% mortality to the bruchid population. The survival bruchid individuals were kept on the VC6089A seeds consecutively for five generations, and it revealed that the mortality of bruchid larvae remained higher than 96%; however, female adults maintained higher fecundity with generations. The toxic-related protein VrD1 purified from VC6089A seed also showed marked resistance to *C. maculatus* under artificial seed bioassays. Thorough inhibition of development was observed when larvae were fed on artificial seeds containing merely 0.2% VrD1, which could be concluded it was one of the most potent anti-bruchid protein discovered from plant so far. Our findings demonstrated the insecticidal activity of VC6089A seeds and VrD1 protein against *C. maculatus*. These results may facilitate pest control corresponding to bruchid infestation, but the potential of bruchids evolving resistance to the resistant mungbean variety through maintaining higher fecundity should be carefully monitored as well.

Productivity and factors associated with arthropod pest abundance in a 2-Year survey of watermelon (*Citrullus lanatus*) production

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We conducted a statewide survey of watermelon production during 1998 and 1999 with 39 and 62 fields (or growers), respectively. The striped cucumber beetle, the squash bug, the melon aphid, and the spider mite were monitored weekly during the growing season. A pest index was developed based on mean densities of these 4 arthropod pests. Among 25 other variables surveyed, plant density, row bedding, presence of windbreaks, hoeing, cultivation rate, rainfall, planting date, and harvest date were strongly associated with this index. High cultivation rates were accompanied with greater arthropod pest abundance. Early planting dates were affiliated with greater abundance of arthropod pests and weeds. However, greater arthropod pest abundance was also correlated with greater productivity in terms of harvest frequency and number of marketable fruit. We partitioned 4 economic categories as machinery, labor, supply, and risk for each survey variable, and then assigned weight scores to each. Management intensity was measured by summing weight scores across the categories for all factors involved in each field and found to be the key for increased productivity. Differences in climatic conditions and sample size may explain the variation in association of factors with pest index and watermelon yield between years.

Effects of vetiver oil and nootkatone on *Myzus persicae* infesting eggplants in a controlled environment

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The essential oil of vetiver (*Vetiveria zizanioides* (L.) Nash.) and the sesquiterpenoid nootkatone, commonly used in the perfume and flavour industries, were reported to show repellent, feeding deterrent and toxic effects to *Coptotermes formosanus* Shiraki, one of major pests of structural wood. The present investigation aimed to study the effects of the two substances on a serious pest of agricultural systems, the green peach aphid *Myzus persicae* (Sulzer), which was maintained in a controlled environment on eggplants (*Solanum melongena* L.), representing one of its most important herbaceous hosts. Preliminary tests were performed to verify the phytotoxicity of formulations of the two substances, using fluorescence microscopy. The activity on *M. persicae* was tested by means of different experiments: a) using foliar disks, and, b) spraying the plants before and after aphid colonization. Evaluations of aphid behaviour and observations on the number of colonizing individuals and of nymphs generated parthenogenetically were performed. The results suggest that, in spite of the problems emerged with the used formulations, both substances could represent potentially good candidates as active ingredients for the control of this aphid.

Effects of spurge flax (*Daphne gnidium* L., Thymelaeaceae) extracts on the development and mortality of the elm leaf beetle (Coleoptera: Chrysomelidae)

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Spurge flax (*Daphne gnidium* L., Thymelaeaceae) is an evergreen shrub typical of the Mediterranean flora. It has been traditionally used for centuries in Spain as a control agent of invertebrate pests of crops and cattle. Previuos bioassays performed in our laboratory demonstrated that leaves and ethanolic extracts of *D. gnidium* showed antifeedant activity on pest species belonging to the orders Heteroptera, Isoptera, Lepidoptera and Coleoptera. The elm leaf beetle, *Galerucella luteola* Müller (Coleoptera, Chrysomelidae), in Spain represents one of the most important pests of elms (*Ulmus spp.*) in both river bank forests and urban areas, being responsible for partial or total defoliation of the tree tops. In order to study the effects of spurge flax on *G. luteola*, different doses of an ethanolic extract of *D. gnidium* were added to the natural diet (elm leaves) of young larvae. During the development, weight changes, phase durations and mortality rates of each stage were recorded. Results showed that the addition of spurge flax extract to the diet of *G. luteola* induced: a) a significant increase in the duration of the larval period; b) greater mortality rates for all stages; c) weight reduction of individuals of all stages, including imagoes.

Termites in the vines: *Kalotermes flavicollis* is one of the main pests of sherry vineyards in Andalusia, Spain

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Sherry vineyards, located in Andalusia (SW Spain) yield important registered designation of origin wines. These vines are affected by important sanitary problems caused by the termite *Kalotermes flavicollis* (Isoptera: Kalotermitidae) and the fungus *Eutypa lata.* A detailed investigation was performed during 15 months in nine districts of the Sherry area on differently aged vines of the "Palomino fino" variety. Samples were collected twice a month, cutting randomly selected vines into six sections, which were examined to detect the living status and the presence of arthropods (*K. flavicollis, Reticulitermes lucifugus*, ants) and fungi (*Eutypa* dieback disease, wood fungi). The results showed that among the 44% unhealthy vines, always aged more than 15 years old, *K. flavicollis*, found in almost 84% of cases, and often in contemporary presence with dead vine areas, represented the major pest of sherry vineyards, followed by *Eutypa* symptoms (14%), whereas *Reticulitermes lucifugus*, ants and wood fungi were found only sporadically. The characterization of the infestations, their trend over the seasons and their location in different sectors of the vines will be shown and discussed.

Direct and correlated responses to selection for varietal resistance in the whitebacked planthopper, *Sogatella furcifera* (Delphacidae)

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Performance of the whitebacked planthopper *Sogatella furcifera* (Delphacidae) was examined on the resistant rice variety IR-58032-1-1 (IR) carrying the resistance gene *Wbph 1*. Nymphal survivorship on IR was as high as those feeding on susceptible rice varieties. In contrast, adult survivorship within 5 days after emergence on IR was only 10-25%. To elucidate whether a planthopper biotype that can attack the resistant IR rice variety can develop, artificial selection for varietal resistance on IR was conducted for 5 generations. A control planthopper line was selected on the susceptible rice variety TN1. Adult survivorship on IR was increased to 40% after 5 generations of selection. However, realised heritability was low (h^2=0.098 for females), suggesting that the biotype may not develop rapidly. Selection on the resistant line not only increased adult survivorship directly but also led to a correlated change in honeydew production (a measure of feeding amount) in two ways. The amount of honeydew in the resistant-selected planthopper line increased on IR, but it decreased on TN1. The results suggest that selection for varietal resistance not only improves feeding ability but also decreases the minimum food requirement to survive. The mechanism of varietal resistance in *S. furcifera* will be discussed.

Detecting new and emerging pests of eucalypt plantations in Queensland: the value of systematised forest health surveillance

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Systematised forest health surveillance has been carried out in eucalypt plantations in southeast Queensland since 2000. Surveys are carried out using two main methods - drive through and walk through. When severe problems are detected intensive monitoring of pest damage and their effects on growth in targeted plantations is carried out by establishing medium-term monitoring plots in which crown damage index (CDI) and growth parameters are assessed, and by using the newly established national standard for CDI assessment. These results feed directly into hardwoods research programs. Call outs from plantation managers and operational staff also form part of the surveillance. The disparate nature of the plantation resource in southeast Queensland means that not all plantations can be visited. Since surveys have been carried out a number of new and emerging pests have been detected. These include an eriophyoid mite (*Rhombacus* sp.) causing severe damage to spotted gums, a plate-galling wasp (*Ophelimus* sp.) on western white gum, *Creiiis lituratus* psyllids on Dunn's white gum and leafblister sawfly (*Phylacteophaga* spp.) on a number of hosts. Spatial analysis of the distribution and severity of damage by the eriophyoid mite is illustrated and data on damage caused by the other emerging pests provided.

Susceptibility of upland cotton *G. hirsutum* to silverleaf whitefly colonization compared to the cotton relative, *Gossypium thurberi*

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Three cotton, *Gossypium hirsutum* L., cultivars and the cotton relative, *G. thurberi,* were evaluated in the field, during 2001, 2002 and 2003, for susceptibility to infestation by the silverleaf whitefly, *Bemisia argentifolii* Bellows and Perring, also known as *B. tabaci* biotype B, in Imperial Valley, CA. The upland cotton cultivars included DP 5415, Siokra L23 and Stoneville 474. Entries were evaluated for colonization by silverleaf whitefly as numbers of adult, eggs and nymphs. In the field studies, *G. thurberi* had very low numbers whitefly adults, eggs and nymphs, followed by Siokra L23, DP 5415 and Stoneville 474. In contrast, in greenhouse studies on potted plants, silverleaf whitefly had higher nymphal survival and shorter development time on *G. thurberi* compared to DP 5415. Low field numbers of *B. argentifolii* on *G. thurberi* does not appear to be explained by poor nymphal survival. The stage-specific survival data from the 2001 greenhouse test, the 2002 data on mature field-hardened potted plants, and the 2003 field survival data support the conclusion that *G. thurberi* is a suitable host for nymphal development.

The change of target thrips and their insecticide susceptibility in Japan

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Melon thrips, *Thrips palmi* Karny, invaded Japan in 1978. Neonicotinoid insecticides were highly active on *T. palmi*, and thus neonicotinoids were used as the main insecticides for thrips control. However, when western flower thrips, *Frankliniella occidentalis* (Pergande), invaded Japan in 1990, the effectiveness of neonicotinoid insecticides on that species was low. The main insecticides to control thrips then changed to emamectin benzoate, spinosad and chlorfenapyr because of their effectiveness in controlling *F. occidentals*. Though onion thrips, *thrips tabaci* (Linderman), have been known as one of the main pests on Welsh onion and onion from the old days in Japan, they were largely ignored owing to the invasion of *T. palmi* and *F. occidentalis.* In recent years, the damage of *T. tabaci* has expanded to other vegetables, fruit trees and flowers, thus raising serious problems in several regions. In order to establish control methods on *T. tabaci*, insecticide susceptibility of *T. tabaci* was investigated. The results showed that the activity of Acephate and Spinosad was outstanding. Neonicotinoids and emamectin benzoate were also active on *T. tabaci*. It suggested that the recent expansion of *T. tabaci* has been caused by several factors beyond just the change of insecticides used on thrips.

New aphid pest of seashore Paspalum turf grass: greenbug, *Schizaphis graminum*

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Greenbug (*Schizaphis graminum*) was first found feeding and damaging seashore Paspalum (*P. vaginatum*) turf grass in October 2003 in Belle Glade, Florida, USA. Feeding by this aphid causes rapid chlorosis and necrosis. Water soaked green spots appear on leaves within 24 h of feeding, followed within 24 hours by yellowing. Leaf death follows within 48 to 72 hours. The use of this turf grass, made popular by its rapid growth, fine short blades and salt tolerance, is increasing internationally in coastal temperate through tropical zones. Greenbug is spreading into golf courses and sod farms in southern Florida and a currently distribution map will be presented. Results of biotype, insecticide efficacy and host plant resistance trials will be shown and discussed in relation to short and long term pest management goals.

The monitoring activity of *Diabrotica virgifera virgifera*, in center part of Romania

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In the maize crop from Romania, about 20 pest species with a great economical importance were identified. In Transylvania, center part of Romania, at maize crop there are recorded pest species on seed and during vegetation, which could induce significant damages by their attack, causing big yield losses and crop compromising. Since 1996, western corn rootworm (*Diabrotica virgifera virgifera)* was registered in western part of Romania, and later, in 2002, in the center part. The monitoring was performed by using feromone sexual traps. Based on the obtained data during these two years of experimentation, the flying curve has been established. In 2002, only six adults were recorded but in 2003, the abundance of adults increased over 400 individuals. The massive appearance of adults (50-60 adults/week) was recorded during flowering stage (July, 15- August, 15). At the present, in Romania, western corn rootworm is identified in 17 districts from 41, but the level of the population does not assure damage area yet.

Wireworms management by seed treatment in Romania

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In Romania, the main species of *Elateridae* are: *Agriotes ustulatus* (40.1%), *A. obscurus* (17.3%), *A. sputator* (11.3%), *A. flavicornis* (10.9%), *A. pilosus* (6.9%), *A. lineatus* (3.8%), *Limonius pilosus* (1.8%), *Synaptus filiformis* (1.6%), *Selatosomus latus* (1.5%). Annually, the wireworms attack 800,000 ha approximately. The recorded densities in field crops vary from 5-10 larvae/sq m up to 70-100 larvae/sq m which determine attack levels up to 25-30% in wheat and barley crops and up to 40-65% in maize and sunflower crops. At present, the seed chemical treatment method is the most important, efficacy, economic and little pollutant measure for the control of wireworms in field crops. At the spring row-crops (maize and sunflower), different products based on the following active ingredients: *acetamiprid, bifentrin, carbofuran, fipronyl, furatiocarb, imidachlopride, thiamethoxam,* have been used. At the winter spiked cereals, the seed treatment was made with products based on *lindan* and one or more fungicides (*carbendazim, carboxine, diniconazol, miclobuthanil, prochloraz, tebuconazol, tiophanat methyl, thiram, triadimenol*). In this case, a good simultaneous protection to both wireworms and some specific pathogens (*Tilletia spp., Fusarium spp., Ustilago nuda, Pyrenophora graminea*) was assured. The efficacy, up to 80% permitted their registration and promoting in production in Romania.

Methods to increase efficiency of releasing *Phytoseiulus persimilis* Athias-Henriot (Acari: Phytoseiidae) predators of twospotted spider mites (*Tetranychus urticae* Koch (Acari: Tetranychidae)) in strawberry

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Florida produces about 2,800 ha. of strawberries for wintertime, fresh market sales valued at about $170 million. Numerous arthropod pests affect the production including strawberry root aphid, melon aphid, fall armyworm, southern armyworm, flower thrips, sap beetles, and *osophila* and fruit flies. No arthropod pest of Florida's strawberry production is more damaging, however, than the twospotted spider mite, *Tetranychus urticae* Koch (Acari: Tetranychidae). Strawberry producers control this pest by collecting weekly ecological information through scouting and applying miticides or *Phytoseiulus persimilis* (Acari: Phytoseiidae) predatory mites at predetermined thresholds. The predators cost about $300/ha. and are shipped in mixtures with dry carriers and are applied by on-farm, hand labor. Experiments were performed to maximize the efficiency of and probability of successful predator releases by the laborers. These data lead to the conclusions that laborers must remix predators and dry carrier material in the shipping containers frequently in order to avoid inefficient releases.

Innovative farm practices for sustainable white grub management in sugarcane

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An integrated management approach was suggested for sustained reduction in grub population of *Leucopholis irrorata* Chevrolat and its damage to sugarcane in the Philippines. The strategy calls for conservation of natural biological control agents through field population enhancement of native grub Scoliid parasites and predators and potential use of *Beauveria bassiana* fungus. Removal of alternate weed hosts, exposure of grubs to solar radiation and predators by thorough soil cultivation, and flooding infested fields reduced grub population. Area-wide collection and burning of collected beetles from harboring trees, before the beetles breed, significantly reduced grub population and the cost of control. Baits and traps using indigenous materials augmented and facilitated beetle collection from large trees. Judicious use of insecticides registered for sugarcane in exceptional cases. Regular monitoring and sustained application of any of these control options, in conjunction with area-wide beetle collection, effectively reduced grub population in infested areas.

Large-scale biological control of cone pests in spruce seed orchards

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We have investigated large-scale application of the biological insecticide *Bacillus thuringiensis* var. k*urstaki*/ *aizawai* (Btk) on damage levels of three lepidopteran species (*Dioryctria abietella* Den et. Schiff. (Pyralidae), *Eupithecia abietaria* Götze, (Geometridae) and *Cydia strobilella* (L.) (Tortricidae)) in a Swedish spruce (*Picea abies* L. (Karst.) seed orchard. To discover the number and timing of sprayings as well as dosage of Btk necessary for control of the lepidopterans, spraying was conducted at different phases of flowering and cone development. The Btk treatment reduced damages caused by *D. abietella* and *Eupithecia* spp. by 65% of that in control treatments. The damage by *C. strobilella* was however not affected by the treatment. Dosage and repeated spraying had no effect on insect damage levels.

Small-scale industrial production of Thai neem-based extracts: RIT-pilot plant and contribution in vegetable insect pests control in Thailand

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By moving-bed contacting extraction technique, defatted cake of Thai neem seeds (*Azadirachta indica* var. *siamensis* (Valeton)) was extracted with methanol at RIT-Pilot Plant (Rajamangala Institute of Technology) in Patumtani province, central part of Thailand. After sedimentation of crude cake, the neem solution was ained out, then filtered and evaporated until a specific volume, the so-called-concentrated alcoholic neem-based extract. Before packing in containers, the content of azadiracthin is monitored per batch by using HPLC. Furthermore the concentrate is also formulated for specific purpose as 3 different "Ready-To-Use" products named- RITNEEM, RITNEEM-O I, RITNEEM-O II. The capacity of pilot plant is 25 ton/year within 250 working days. In contribution to vegetable production, many field experiments on insecticidal toxicity against *Hellula undalis, Lipaphis erysimi, Spodoptera litura* and *Pleutella xylostella* in chinese kale were conducted. Overall, it was observed that similar trend of population suppression were noted between all formulations at dose of 25 cc/20 L. In particular, they could be considered to give effective control against *H. undalis* and *S. litura*, whereas *P. xylostella* was the least susceptible. However, all formulations did not result in significantly greater yields than control with chemical and water spraying.

Non-host anti-feedants in *Hylobius* pine weevils: from lab to field applications

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Anti-feedants as a protection against herbivores is known in many plants, like in the 'neem' tree *Azadiractica indica.* In conifers, defence is quantitative, based on less toxic oleoresin compounds. In small, stressed conifers like the ones planted on clear-cuts in forestry, the quantitative defence is weak. These seedlings have been defended by insecticides like DDT and pyrethroids against the feeding by adult pine weevils *Hylobius abietis* but use of insecticides will be banned. Our work follows two lines: 1) The screening of synthetically available anti-feedant compounds and related structures. 2) The search for antifeedants in non-host plants (presented elsewhere). The development of laboratory bioassays has been a prerequisite for both. The screening of synthetic antifeedant candidates has given a dozen field test candidates. Three groups of interest, the carvone-analogues, the arylaldehydes, and the bark alkaloids, show a high activity for some structures. Field-tests in 1997 and 2001-2003 have confirmed the activity in lab-bioassays, but the molecular activity itself is here interwoven with effects of stability and evaporation. The three factors compound, dose, and formulation and all affect protection efficacy. New types of formulations in development, like thicker coatings or discrete or provoked-release principles show promising first results.

Sweetpotato weevil, *Cylas formicarius elegantulus* (Summer) (Coleoptera: Curculionidae): a worldwide pest of sweetpotato and sustainable management

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Sweetpotato weevil (SPW) is the most destructive insect pest of sweetpotato in the world. Native to Asia, SPW was discovered in India in 1792 and in US in 1875, near New Orleans, LA. Occurrence on five continents and 11 US southern states attests to weevil's adaptive potential and impact. Weevil dissemination has historical ties to people and commerce. Sweetpotatoes, *Ipomoea batattas*, and related yams, *Dioscorea* sp., are the seventh largest crop (FAO world crop statistics). Weevil hosts include seaside morning glory, *Ipomoea sagittata* and related species. Sweetpotato production and marketing offer constant weevil spread. Direct damage by this quarantined pest, control costs and regulatory action conservatively exceed $20 M annually in USA. SPW management is sustained through an educational program, through development and worldwide use of sex pheromone trapping, tolerant varieties, effective insecticides, sound cultural practices, micropropagation techniques and regulatory programs. To contain weevil movement and control or successfully manage weevils requires cooperation, synchronization and application of any and all IPM tactics and strategies. Research efforts with breeding, DNA finger printing in combination with host plant resistance, pathways of movement and detection, insecticidal control in field and storage, cultural and biological control, in concert with eradication and educational programs should improve understanding of SPW behavior, ecology and management.

A tractor mounted device for collecting live arthropods in cotton crops.

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Efficient and precise arthropod sampling methods are important components of integrated pest management in Australian cotton crops. Here we describe a tractor-mounted device for sampling and collecting live insects and spiders from cotton crops, and we present the results of a single experiment quantifying the effectiveness and precision of the device. The number and diversity of insects and spiders captured per metre of row by the device were significantly lower than the average results from beat-sheets that sample individual metres. However the device can sample much greater distances per unit time. The total number of individual arthropods captured by the device increased as tractor-run length increased from 20 to 40 and 80 metres. However the increase was not linear, and variance increased substantially with run length. The total number of distinct taxa collected peaked at tractor-run lengths of 40 metres. Overall the device is less efficient and less precise than manual beat-sheet sampling for the purposes of crop protection decisions. However the device does provide an efficient and effective method for the collection of live insects and spiders from cotton crops.

Advances in IPM for citrus in New Zealand

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The citrus industry in New Zealand is relatively small (approximately 2000 ha) compared to other New Zealand grown fruit crops such as apples (approx. 14200ha) and kiwifruit (approx. 11500ha). Because of the small size, the citrus industry has historically received little research attention and has fallen behind with respect to IPM developments in other crops. However, in 2002, a three-year programme was initiated to develop an IPM programme for New Zealand citrus. The citrus IPM programme builds on past experiences gained from other IPM programmes developed in New Zealand, especially those developed for pipfruit and kiwifruit. Successful models for analysis of industry data have been applied to citrus to determine current industry practices and monitor changes over time. Knowledge of successful IPM approaches for generic horticultural pests and diseases have also been included. However, many of the pests and diseases are unique to citrus and there is a limited ability to easily transfer technology and approaches from one crop to another. For example, thrips, especially *Pezothrips kellyanus*, is one of the key pests iving the pest management programme in citrus, whereas thrips are not a major pest of kiwifruit or pipfruit. Specific research to identify IPM-compatible methods for controlling thrips, scale insects and mites is underway. Determining the role of grower practices on natural enemies is a key component of the project.

Dependence of biotype development on life history traits of pests

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Biotype management is crucial for both the breeding and sustainable use of pest-resistant crops. We developed simulation models accounting for a wide range of pests to evaluate the effects of life history traits on the developmental velocity of a new biotype. Parameters such as the proportion of land cultivated with a resistant variety, migration rate among patches, reproductive potential and degree of density dependence in population growth, initial frequency and dominance of a biotype gene, and fitness cost of a biotype on susceptible varieties were incorporated into 1-locus 2-allele models. The simulation results revealed that life history traits promoting assortive mating have prominent effects on biotype development in general. The frequency of a biotype gene remains at a negligible level for more than 25 generations in holometabolous pests that mate after migration, provided that the biotype gene is recessive and less than 0.1% in initial frequency. Migration rate does not greatly affect biotype development, while high reproductive potential and strong density dependence in population growth of pests facilitates biotype development. On these results we discuss desirable strategies for breeding durable resistant varieties.

Ripening stages of rice spikelets selectively damaged by four species of rice bugs, *Leptocorisa chinensis* Dallas (Hemiptera: Alydidae), *Lagynotomus elongatus* (Dallas) (Hemiptera: Pentatomidae), *Cletus punctiger* (Dallas) (Hemiptera: Coreidae), and *Stenotus rubrovittatus* (Matsumura) (Hemiptera: Miridae).

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The rice bugs *Leptocorisa chinensis*, *Lagynotomus elongatus*, *Cletus punctiger*, and *Stenotus rubrovittatus* are serious economic pests of rice crops in Japan. To clarify the ripening stages of spikelets preferentially attacked by those bugs in paddy fields, rice bugs were individually released for three days onto a rice panicle at the following stages of maturation: 7, 14, 21, and 28 days after heading. The spikelets were classified into three groups according to the flowering order in a panicle. The spikelets in Group A (early flowering) and B (mid flowering) were selected by the bugs to feed on at 7 DAH; that is, earlier than those in Group C (late flowering). At 14 and 21 DAH, however, bugs preferred the spikelets in Group C to those in the other two groups. In *L. chinensis*, *L. elongatus*, and *S. rubrovittatus*, the total number of damaged spikelets decreased with maturing of the panicle, whereas there was no remarkable change in the number of spikelets damaged by *C. punctiger*. These results suggested that all species of rice bugs except *C. punctiger* selectively damage the spikelets whose ovaries are developing from lengthwise to sidewise elongation, whereas *C. punctiger* mainly attacks the spikelets whose ovaries are developing from lengthwise to thickness elongation.

WideStrike™ a stacked Bt cotton for improved lepidopteran pest control and resistance management

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Dow AgroSciences is a major supplier of pest management, biotechnology and seed products that improve the quality and quantity of the earth's food supply and contribute to the health and quality of life of the world's growing population. One of the more prominent developments in insect control has been the development and rapid adoption of Bt cotton which brought significant advantages to the cotton producer. Dow AgroSciences has had a long commitment to biotechnology which is bearing fruit with a highly efficacious stacked transgenic trait that express both the Cry1F and Cry1A lepidopteran active Bt proteins in cotton plants. The combined trait stack results in higher levels of efficacy and a broader lepidopteran pest spectrum than previously available in single Bt cotton improving the overall value to the grower and providing selection diversity to slow resistance development in target insects. Additionally, improved promoters provide expression throughout the growing season. Dow AgroSciences anticipates that WideStrike will be available commercially in the U.S. via PhytoGen cotton varieties in 2005. Dow AgroSciences is supporting licensing of WideStrike to additional seed companies and geographies where all environmental safety and stewardship requirements can be met. Performance data and information on pest spectrum will be presented.

Impact of artificial shelters on arthropod complex in agroecosystem

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Artificial shelters were made of straw as long banks in large fields of winter wheat. The most pronounced positive effect was observed in active predators using these shelters. Abundance of some beetles (*Carabus*, aleocharine staphylinids) and Lycosidae spiders were increased in some times in close vicinity to the shelters. The same increase takes place in herpetobiont beetles Anthicidae which also hid under straw. Other mass herpetobiont beetles of Dermestidae and Silphidae did not use these shelters. The abundance of *Dermestes laniarius* does not depend on the presence of shelters while the beetles of *Silpha obscura* avoided the field zones with shelters. Parasites and herbivores were observed in shelters very seldom. Catches of hortobionts (aphids, bugs, thripses and hymenopterans) by entomological net as well as the parasitization of eggs of a harmful bug, *Eurygaster integriceps*, were not influenced by shelters. However the number of eggs that yielded neither bug larvae nor their parasites (killed by predators?) increased twice as many at the parts of field with shelters.

Imidacloprid treatment of seedling roots as a component of an integrated pest management strategy for insect pests in *Brassica* crops.

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In Queensland, Australia *Brassica* crops are attacked by a suite of insect pests. In the early months of the growing season (February- April), crops are attacked by *Bemisia tabaci* type B and the major lepidopterous pests are *Hellula hydralis* and *Crocidolomia pavonana*. Later in the season (May- September) the diamondback moth, *Plutella xylostella*, poses the greatest threat to the successful cultivation of *Brassica* crops. Disruption of the agro-ecosystem by the frequent foliar application of synthetic chemical insecticides to control these early season pests can seriously diminish the success of subsequent integrated pest management programmes developed for the diamondback moth. In field studies, root treatment of early season cabbage seedlings with imidacloprid prior to transplanting significantly reduced *H. hydralis* infestation for up to four weeks post transplantation and *B. tabaci* infestation for up to five weeks after transplantation. *Crocidolomia pavonana* infestation was not affected by imidacloprid treatment of seedlings. Root treatment of seedlings with imidacloprid can reduce the number of foliar sprays required for the management of some early season *Brassica* pests and has the potential to be incorporated into existing pest management programmes without disrupting the successful management of the diamondback moth.

Yearly fluctuations in the immigrant density of the rice planthoppers into Japan

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Since the mid 1990s, the immigrant densities of the migratory planthoppers, *Sogatella furcifera* and *Nilaparvata lugens* from south China to Japan have been decreased compared with those in the 1980s. Light trap catches obtained in southern part of Japan in June and July were used for detailed analyses. No correlation was observed in the number of catches between *N. lugens* and *S. furcifera* for the period from 1997 to 2002, whereas high correlation was observed in the trap catches between *N. lugens* and *S. furcifera* during 1989 to 1993. Only 5 to 20% of the total catches of *S. furcifera* were obtained in June early in the 1990's, but the proportion increased 30 to 70% during 1997 to 2002. The mean cumulative days of low-level jet stream (LLJ) development observed during June and July decreased from 15.0 days/year in 1980-1990 to 8.4 days/year in 1997-2002. The differences between the two periods was due to much lower frequency of LLJ development in July in 1997-2002. This indicates that the absence of meteorological conditions for migration accounts for the recent decrease in immigrant planthoppers in Japan, though low population densities in the migrant source area is another possible reason.

No adverse effect of YieldGard® rootworm on ground beetles (Coleoptera: Carabidae)

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The effect of transgenic corn ground beetles was studied under laboratory and field conditions. No differences in ground beetle populations occurred in transgenic and non-transgenic corn in the field. No differences in longevity and fecundity occurred in laboratory tests where two ground beetle species were fed dog food sprinkled with pollen from transgenic and non-transgenic plants.

Spatial and temporal features and social composition of vine-infesting colonies of Kalotermes flavicollis in Sherry vineyards (Spain)

MA Lopez, R Ocete, L Maistrello

Paper not available at time of production

Predators for the control of spider mites in Japanese apple orchards

S Toyoshima

Paper not available at time of production

Reproductive incompatibility and differences in host range between the deciduous-tree and evergreen-tree strains of Oligonychus gotohi (Acari: Tetranychidae)

S Abe, T Gotoh

Paper not available at time of production

Status of three invasive insects recently established in Hawaii

AH Hara, CM Jacobsen, EM Domingo

Paper not available at time of production

Recent invasion of Japan by some gall midges (Diptera: Cecidomyiidae) of economic importance

M Tokuda, N Ueichi, J Yukawa, F Kawamura

Paper not available at time of production

Research on the environmental impacts of GM plants in New Zealand

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In 2001 New Zealand's Royal Commission on Genetic Modification (GM) gave a recommendation to "proceed with caution" and to undertake more research on the potential environmental impacts of this technology. Studies to develop methods in containment for assessing impacts on non-target organisms (bees, predators and soil biota) were already underway. This effort was expanded with new research programmes on horizontal gene transfer in NZ soils, gene flow to indigenous and naturalized plants, native pollinators of crop plants, persistence of gene products (recombinant proteins) in soil and in insect frass, criteria for selecting non-target invertebrate species for biosafety testing, and specific environmental impact studies with GM vegetable crops and forest trees. Recent results from a honey bee project suggest that three different insecticidal gene products (a Bt toxin, a proteinase inhibitor and a biotin-binding protein) are not likely to affect the development of hypopharyngeal glands in adult bees that have consumed each protein with their pollen food. Recent results from a project to examine the effects of GM plants on soil biota suggest that proteinase inhibitor-expressing plants have little impact on life cycle parameters in one species of Collembola.

The Silkworm Genome Project in China: a 6X draft sequence of the domesticated silkworm (Bombyx mori) genome

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In 2003, the international silkworm genome project Chinese group has finished a 6x coverage sequencing of the domesticated silkworm *(Bombyx mori),* by using of whole genome shotgun (WGS) technique. Here we report a draft sequence for the genome of the domesticated silkworm, *Bombyx mori*. Our assembly contains 92.9% of all known silkworm genes. The gene number of silkworm genome is estimated to be 18,510, more than that of *Drosophila melanogaster*. Comparisons to the other two sequenced insect genomes**,** fruitfly and mosquito, reveal some remarkable parallels and differences. We reported the main strategy and results of this project as following: 1) The strategy of sequencing 2) The general results of sequencing 3) Assembly of the silkworm genome sequences 4) Annotation of the silkworm genome 5) Genomic comparison of silkworm and other insects 6) Biological analysis of the silkworm genome 7) Outline of the further study in China

Population genetics of kdr-type resistance to pyrethroids in the peach-potato aphid, *Myzus persicae***.**

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The peach-potato aphid, *Myzus persicae,* is an important agricultural pest in many parts of the world. It is mainly controlled through insecticide applications and has developed strong resistance to most of the major chemical classes as a result of their widespread use. Resistance to pyrethroids is particularly problematic in *M. persicae* and is primarily the result of a target site resistance mechanism termed knockdown resistance (or kdr). We have found that this is caused by either one (L1014F) or two (L1014F + M918T) point mutations in the domain II region of the para-type voltage-gated sodium channel (the target for pyrethroids). Aphids that carry the double mutation are highly resistant to even the most potent pyrethroids, surviving doses of deltamethrin in excess of 100ppm. To facilitate rapid monitoring for pyrethroid resistance in *M. persicae*, we have developed an allelic discrimination PCR assay to determine the genotypes of the two mutations in individual adults using fluorescent Taqman® MGB probes. We are using this assay to study the origins and spread of the mutations in a diverse range of *M. persicae* populations.

Insecticide resistance and its management in the cotton bollworm *Helicoverpa armigera* in Pakistan

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The cotton bollworm, *Helicoverpa armigera* (Hubner) (Lepidoptera: Noctuidae) is a key pest of cotton and other crops in Pakistan. Indiscriminate use of pesticides prompted the development of insecticide resistance in this pest. Resistance to pyrethroids has been on the rise since 1991. Resistance factors are now >100 to all the pyrethroids. Endosulfan resistance remained moderate to high. However, some thiophosphates and carbamates mostly exhibited low levels of resistance. We never ran out of options for the control of *H. armigera*. The latest introduction of new chemistries with novel modes of action added to the effective armory. An IRM strategy including other IPM tactics has been recommended to the cotton growers for the better control of *H. armigera* and other cotton pests.

Mutations in acetylcholinesterase (AChE) of the cotton melon aphid (*Aphis gossypii* Glover) and the peach potato aphid (*Myzus persicae* Sulzer) confer resistance.

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Two mutations in the *ace1* gene of *Aphis gossypii* are associated with insensitivity of acetylcholinesterase (AChE) to carbamate and organophosphate insecticides. The first of these, S431F (331 in *Torpedo californica* numbering), is associated with insensitivity to the insecticide, pirimicarb. The mutation to a phenlyalanine is consistent with a loss of sensitivity to pirimicarb in a range of both *A. gossypii* and *M. persicae* clones. A rapid RFLP diagnostic allows the identification of individuals of both aphid species with a resistant genotype. This diagnostic further reveals the presence of S431 in several other pirimicarb-susceptible aphid species. The serine at this position in the wild-type enzyme has only been reported for aphids and is considered the reason why pirimicarb has a specific aphicidal action. A second mutation, A302S (201 in *T. californica*), confers insensitivity to a wide range of carbamates and organophosphates and has, to date, only been found in resistant strains of *A. gossypii.*

Stem application of monocrotophos in okra: dissipation of monocrotophos residues

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The status of residues of monocrotophos at harvest and its dissipation pattern in okra fruits when employed as stem application at the rate of 1:6 (one part of monocrotophos in six parts of water) were studied in a field experiment. The residues of monocrotophos were below detectable level (BDL) during first picking when the stem application was imposed at or before 38 days after germination (DAG). The repeated application, even during fruiting, evidently established the cumulative residues of monocrotophos in okra fruits. The residues (0.57 ppm) recorded in okra fruits on first day of stem application accumulated up to 3rd day (2.21 ppm) and dissipated gradually to maximum residue level (MRL) of 0.2 ppm with Ttol. of 11.27 days. Therefore stem application of monocrotophos in okra should be restricted to 12 days prior to picking of fruits.

Effects of cypermethrin and chlorpyrifos residues in cattle dung on the development of the dung beetle *Copris tripartitus* (Coleoptera: Scarabaeidae)

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Fresh dung was collected from untreated cattle and cattle dosed with a spray-on formulation of cypermethrin and chlorpyrifos (2.1g /cow) on days 1, 3, 5 and 7 post-treatment. The dung was bioassayed using the dung beetle *Copris tripartitus* Waterhouse. Residues of cypermethrin and chlorpyrifos were sufficient to inhibit oviposition by *C. tripartitus* in day 1 dung but there was no significant effect on egg laying in dung collected at days 3-7 post-treatment in sexually matured female. But, in the second generation residues of cypermethrin and chlorpyrifos were sufficient to inhibit oviposition by *C. tripartitus* in treatment after 1 and 3 days. Although there was no significantly different effect on egg laying in dung collected at days 3-7 post-treatment in first breeding but, in second breeding, toxic effect reveal in 3 and 5 days dung treatments. Nulliparous dung beetles fed for 200days on dung collected 0, 1 and 3 days after treatment of cow with cypermethrin and chlorpyrifos show little ovarian development.

Effect of ecdysone agonists on tortricid male/female orientation

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The attractiveness and responsiveness of adult codling moth, *Cydia pomonella* (L.), redbanded leafroller, *Argyrotaenia velutinana* (Walker), and obliquebanded leafroller, *Choristoneura rosaceana* (Harris), exposed to surfaces treated with the ecdysteroid agonist methoxyfenozide were investigated in wind tunnel and orientation tube assays. When males were exposed to water-treated surfaces for 48 h the mean percent recaptures of such treated males in the wind tunnel assay were almost always (depending on species) significantly greater than the recaptures of males that had been exposed to methoxyfenozide-treated surfaces. Similarly, in the orientation tube assay, methoxyfenozide-treated males almost always had significantly lower mean levels of individuals exhibiting sexual excitability (intense wing fanning and upwind orientation) and mean distances traveled upwind in the orientation tubes. The two assays demonstrated that males exposed to methoxyfenozide-treated surfaces were not as responsive to calling females (treated and non-treated) as were the non-treated males. Conversely, females exposed to methoxyfenozide-treated surfaces were just as attractive to non-treated males as were water-treated females. It appears that a male's ability to respond to a calling female is more negatively affected by the ecdysone agonist than a female's ability to call and attract males.

Assessing the role of cytochrome P450s in insecticide resistance of *Lucilia cuprina*, *Helicoverpa armigera*, and *Drosophila melanogaster*

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Cytochrome P450 are heme containing enzymes that perform a diverse array of REDOX reactions against exogenous and endogenous compounds. Characterised insect genomes have approximately 90-100 individual cytochrome P450 genes and the biological function for most of these is unknown. Previous studies have implicated the over-expression of some cytochrome P450 genes in insecticide resistance via metabolic degradation. Using degenerate PCR and cDNA library sequencing, we have cloned and characterised a number cytochrome P450 genes from the Australian sheep blowfly (*Lucilia cuprina*) and the cotton bollworm (*Helicoverpa armigera*), two insect pests of Australia. We have generated a micro-array of these cytochrome P450 genes, and are characterising the expression of these genes at different life cycle stages and in different tissues. We are also using - micro-array and realtime quantitative PCR to compare the expression levels of cytochrome P450 genes between resistant and susceptible strains. Our studies in *L. cuprina* and *H. armigera* are complemented by studies in *osophila melanogaster*. Using a micro-array containing all 90 *D. melanogaster* cytochrome P450 genes, we are studying gene induction by insecticides. We are over-expressing induced cytochrome P450 genes, and determining if over-expression can confer insecticide resistance.

Novel genetic basis of field-evolved insect resistance to Bt toxins

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Insecticidal toxins from *Bacillus thuringiensis* (Bt) are widely used in sprays and transgenic plants to control insects that attack crops and transmit diseases, but evolution of pest resistance threatens their continued efficacy. Diamondback moth (*Plutella xylostella*) is the only pest that has evolved Bt resistance in open field populations, although several others have been selected for resistance in the laboratory or glasshouses. The most common type of resistance (Mode 1) is characterized by recessive inheritance, >500-fold resistance to at least one Cry1A toxin, and negligible cross-resistance to Cry1C. The primary mechanism of Mode 1 resistance is reduced binding of Bt toxins to midgut membrane target sites. Mutations affecting a Cry1A-binding midgut cadherin protein are tightly linked to laboratory-selected Mode 1 resistance in two major pests, *Heliothis virescens* and *Pectinophora gossypiella*. The cadherin gene has thus been considered the prime target for DNA-based screening for resistance. Here we show that field-evolved Mode 1 resistance in diamondback moth has a different genetic basis, indicating that screening for resistance in the field should not be restricted to searching for cadherin mutations.

Managing pest resistance to Bt crops in India

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Bt transgenic cotton first introduced in India in 2001 was cultivated on over 100,000 hactares in 2003. Besides, other Bt transgenic crops such as rice, egg plant, cole crops, pulses are in various stages of development. The target pest for Bt cotton is the American bollworm, *Helicoverpa armigera* and for Bt rice it is yellow stem borer (YSB), *Scirpophaga incertulas*. Based on the baseline toxicity data against the former in India and estimation of frequency of resistance alleles in the populations of YSB in the Philippines, it appears that these two species can rapidly buildup resistance against the Bt endotoxins. Bt resistance management for transgenic cotton is made mandatory requirement for the growers who have to maintain 20% refuge crop or minimum of 5 rows around the Bt cotton field, whichever is larger. However, in view of inadequate knowledge and expertise among the farmers, small land holdings and apprehension of expected reduction in yield due to non Bt lines, farmers tend to ignore the refuge cultivation. Studies on the baseline susceptibility of the American bollworm to Cry1Ac have helped in the monitoring of resistance in the Bt cotton. Similar studies are being done in other crops too. We feel, Bt resistance management should include detailed studies on various characteristics of Bt resistance in local pest populations of the target pests, and explore possibilities of field level management in tandem with the local agronomic practices and environmental conditions.

Bt maize effects on butterflies.

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Transgenic plants have been widely adopted by U.S. farmers despite concerns about potential nontarget impacts of this technology. The possibility that maize (*Zea mays*) genetically engineered to express *Bacillus thuringiensis* endotoxoin for control of European corn borers (*Ostrinia nubilalis*) may have impacts on non-target lepidopterans through pollen drift onto neighboring non-agricultural plants has received considerable attention. Since initial concerns about Bt corn were raised by a laboratory study, additional studies both in the field and the laboratory have revealed the remarkable complexities of accurately assessing risks of pest management practices in agroecosystems. These studies are examined in the context of ecological, economic, social, and cultural factors that affect food production and pest management in the United States.

The relative fitness of Bt resistant and susceptible *Helicoverpa armigera* – implications for the refuge strategy

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Transgenic crops producing the Cry1Ac toxin of *Bacillus thuringensis* (Bt) was introduced in Australia, China and India primarily for the control of *Helicoverpa* spp. As *Helicoverpa armigera* has a demonstrated capacity for developing resistance to many synthetic insecticides, development of resistance to Bt toxins remains the biggest threat to the sustainability of transgenic technology. The refuge strategy has been widely adopted to counter the onset of resistance. This strategy is most effective when resistance is conferred by rare, recessive alleles and when mating of heterozygotes in prevented. Under these conditions, the evolution of resistance can be substantially delayed. However, we have found evidence that two of these conditions are not always met. Results from glasshouse experiments have demonstrated that, although resistance in *H. armigera* is functionally recessive on young Cry1Ac cotton, attenuation in toxin expression results in survival of heterozygous genotypes on late season Cry1Ac cotton. Our resistant strain of *H. armigera* takes longer to develop on cotton that expresses high levels of Cry1Ac than their susceptible counterparts on non-Bt cotton. However, loss of potency in older cotton plants mitigates asynchrony in late season insect populations. The implications of these findings for resistance management of Bt crops will be discussed.

Novel approaches for controlling mosquitoes using natural inhibitors for trypsin modulating oostatic factor

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Trypsin Modulating Oostatic Factor (TMOF) is a mosquito decapeptide that modulates trypsin biosynthesis in mosquitoes, flies and *Heliothis virescens*. Feeding the hormone and its analogues to mosquito and *H. virescens* larvae stopped trypsin biosynthesis in their midgut by binding a specific gut receptor and controlling the translation of the trypsin message. Cytoimmunochemical analyses show that the hormone is not only synthesized in the ovary as was initially hypothesized, but is also found in the central nervous system cells of adults and larvae. Since the hormone affects the growth and development of mosquito larvae the hormone and several analogues have been cloned and expressed in chlorella, yeast (*Saccharomyces cerevisiae* and *Pichia pastoris*) and Alfalfa cells to control insects using biorational insecticides. These results and the effect of the transformed cells on larval growth and development will be discussed.

Fluorometric PCR applications to insecticide resistance analysis in disease vectors.

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As insect genomes are published, molecular techniques have begun to be widely applied for the detection and assessment of insecticide resistance. Vectors develop insecticide resistance through mutations of insecticide target sites, such as the voltage-gated sodium channel, and increased activity of detoxification enzymes such as oxidases and esterases. The ability to use preserved or desiccated field-collected insects makes molecular analysis of insecticide resistance an especially attractive option. A useful approach to measuring all of these mechanisms is fluorometric PCR. This method allows detection of resistance mechanisms based on point mutations at the insecticide target site, resistance gene amplification, or increased expression of resistance genes. Knowledge of resistance gene sequence is a prerequisite in the development of these methods. Our efforts to apply quantitative PCR to detection, measurement and assessment of insecticide resistance in an assortment of disease vectors are summarized. The same molecular methods that serve to detect resistance are also central to the determination of population genetic relationships of the resistance alleles. We discuss our present state of knowledge of molecular markers and insect genomes as they relate to insecticide resistance.

Tri-trophic prey-mediated impacts of insect-resistant tobacco on a predatory carabid beetle

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Larvae and adults of a New Zealand endemic predatory beetle, *Ctenognathus novaezelandiae*, were fed an exclusive diet of prey (*Spodoptera litura*) reared on control tobacco, or transgenic tobacco expressing either a protease inhibitor (PI) or a biotin-binding protein (BBP). BBP prey-fed beetle larvae accumulated greater mass than the other 2 treatments over a 7-week period in the middle of the 14-week experiment. Over the entire experiment, the mean biomass of BBP-fed larvae was greater than that of PI- but not control-fed larvae. There were no differences among treatments in larval survival or quantity of prey consumed. Adult female beetles grew larger and survived longer than males in all 3 treatments. Combined gender adult mean biomass over 40 weeks was the same for all 3 treatments. However, control-fed females had greater total biomass than PI or BBP-fed females, but control-fed males had less biomass than males given the other treatments. Over the entire experiment, BBP-fed adults consumed slightly less than the other 2 treatments. Fecundity was low in all 3 treatments and there were no differences among them. The greatest adult differences detected were between genders rather than among treatments, where analysis of data collected weekly showed only minor and transient effects.

Analysis of acetylcholinesterase insensitivity in California insect pests

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Acetylcholinesterases are important members of the serine hydrolase super-family of enzymes and are the targets of the organophosphate (OP) and carbamate insecticides. Although the future use of these insecticides in California is somewhat uncertain, due to the FQPA, they are still widely used in agriculture, both as stand-alone insecticides, and as synergists of the pyrethroid insecticides. As a consequence of their widespread and varied usage, OP and carbamate resistance has been identified in many important pests. Acetylcholinesterase insensitivity is an important mechanism of resistance to these insecticides, and cross-resistance within these chemical groups is not uncommon. We have conducted extensive studies on resistance in several important California insect pests. In this presentation, we will describe our studies on two pests – the beet armyworm (BAW), *Spodoptera exigua*, and the glassy-winged sharpshooter (GWSS), *Homalodisca coagulata*. The BAW has been long-established in California agriculture, whereas the GWSS is a recent introduction. These two pests differ in their pesticide exposure histories and this seems to be a contributing factor in their relative susceptibilities to OPs and carbamates. The effectiveness of OPs and carbamates for the management of these pests is discussed based on our toxicological and biochemical studies.

Activity profiles of neonicotinoid insecticides in plants and implications for IPM

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The duration that an insecticide residue is active in a crop in terms of lethal and sub-lethal effects on insect populations is a critical concept in pest management that has been poorly developed. Application rates, the timing and spacing of applications, and the choice of materials could all be influenced by more reliable information on the activity period of insecticides in crops. Commercial ELISA kits were used to determine concentrations of imidacloprid and thiamethoxam in citrus, grapes, and melons at regular intervals following soil treatment. The uptake and residual profiles differed significantly for both compounds in all crops. Peak concentrations of imidacloprid in xylem fluid of large citrus trees were not attained until 6-8 weeks after application compared to 2 weeks for thiamethoxam. However, near-peak levels of imidacloprid were sustained for 6-10 weeks before declining gradually in contrast to thiamethoxam titres (applied at ¼ the rate of imidacloprid) that declined rapidly. The same pattern of higher application rates and longer residual of imidacloprid was also observed in melons. Comparison of young vs. old leaves on primary branches of melon vines revealed much higher concentrations in older leaves, a counterintuitive finding because better whitefly control is attained on younger leaves.

Contamination of refuges by Bt toxin genes from transgenic corn

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Transgenic crops producing insecticidal toxins from *Bacillus thuringiensis* (Bt) are widely used to control pests, but their benefits will be lost if pests evolve resistance. The mandated high-dose/refuge strategy for delaying pest resistance requires planting refuges of toxin-free crops near Bt crops to promote survival of susceptible pests. We report that pollen-mediated gene flow up to 31 m from Bt maize caused low to moderate Bt toxin levels in kernels of non-Bt maize refuge plants. Immunoassays of non-Bt maize sampled from the field showed that the mean concentration of Bt toxin Cry1Ab in kernels and the percentage of kernels with Cry1Ab decreased with distance from Bt maize. The highest mean Bt toxin concentration in kernels of non-Bt maize plants was 45% of the mean concentration in kernels from adjacent Bt plants. Most previous work on gene flow from transgenic crops has emphasized potential effects of transgene movement on wild relatives of crops, lanaces, and organic plantings, while implications for pest resistance have been largely ignored. Variable Bt toxin production in seeds of refuge plants undermines the high-dose/refuge strategy and could accelerate pest resistance to Bt crops. Thus, guidelines should be revised to reduce gene flow between Bt crops and refuges.

Insect comparative genomics: a carboxyl/cholinesterase perspective

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Our comparative genome analyses of principle enzyme families involved in metabolic resistance to insecticides have generated new research opportunities. The data provide a credible basis for identifying the resistance-associated enzymes of insects and the means for identifying previously unknown molecules involved in fundamental biological process such as development. Carboxyl/cholinesterase enzymes have contributed to a major part of insect biochemical and genetic research. The most notable member of this supergene family, acetylcholinesterase (AChE), is the key processing enzyme for the neurotransmitter acetylcholine. Many other esterases have now been implicated in the metabolism of specific hormones and pheromones and accordingly in various aspects of insect development and behaviour. Intriguingly, the carboxyl/cholinesterase family provides a striking example how gene duplication/radiation/deletion shape the evolution of novel biology, notably how the function of a catalytically active domain has evolved to mediate receptor-ligand interactions that regulate morphogenetic process during development of the nervous system. We have recently classified carboxyl/cholinesterases into three principle functional groups (intracellular enzymes associated to dietary detoxification; secreted enzymes including those that process pheromones and hormones; receptors and enzymes involved in neuro/developmental functions). Our data (gene inventories) are also providing a template for new studies concerning the honeybee and silkworm genomes.

Characterization of CTCF proteins in mosquitoes

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CTCF (CTCCC-binding factor) is a multivalent, multifunctional transcription factor well characterized in vertebrates. Of particular interest is its association with every known vertebrate insulator and its emerging role in epigenetic regulation. Endogenous mosquito insulators would be valuable elements to flank transgene insertions, inhibiting both position effect and position effect variegation, thus allowing for more consistent gene expression. CTCF-like cDNAs were cloned, expressed and characterized from the medically important mosquitoes *Aedes aegypti* and *Anopheles gambiae.* The gene is transcribed constitutively in all developmental stages and appears to be up-regulated both in the ovaries from blood-fed females and in early embryos. Putative binding sites for the *An. gambiae* protein were determined *in-vitro* using PCR-assisted binding site selection and confirmed in gel-shift assays. ChIP (chromatin immunoprecipitation) assays are currently in progress. Additionally, an RNAi experiment to knockdown expression both in *An. gambiae* adult females and in *osophila melanogaster* females and early embryos, followed by RNA isolation and hybridization to microarray chips is being performed to identify specific CTCF targets. The results of these experiments will be presented.

Esterase mediated cross resistance between pyriproxyfen and buprofezin in B-biotype *Bemisia tabaci* in Australia

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B-biotype *Bemisia tabaci* was first detected in Australia in 1994. B-biotype *B. tabaci* is a severe pest worldwide and poses a serious threat to many ornamental, agricultural and horticultural industries in Australia. Control of B-biotype *B. tabaci* is hampered by resistance to many conventional insecticides. Consequently, insect growth regulators such as buprofezin and pyriproxyfen are important insecticides in managing this pest. Field resistance has been detected to both insecticides with cross resistance between the two as well. Studies have shown resistance to buprofezin in B-biotype *B. tabaci* to be esterase mediated and experiments were designed to investigate the potential of pyriproxyfen to bind to *B. tabaci* esterases. Esterase inhibition experiments showed inhibition by pyriproxyfen was greater in the buprofezin resistant strain than in its unselected counterpart. Concentrations as low as 0.321nM pyriproxyfen gave 70-80% inhibition of resistant *B. tabaci* esterases. Cross resistance between IGRs with different modes of action poses a grave threat to the viability of the management strategy of silverleaf whitefly in Australia.

Efficacy of novaluron for Colorado potato beetle control and potential for resistance development.

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Novaluron (RimonÒ 10EC), a novel insect growth regulator, could play a key role in development of practical integrated pest management (IPM) programs for Colorado potato beetle (CPB), *Leptinotarsa decemlineata* (Say). In laboratory tests, novaluron exhibited potent insecticidal activity against CPB larvae (e.g. 2nd instar (L2) LC_{50} = 0.42 ppm). Furthermore, the insecticidal activity of a single novaluron application was highly persistent under field conditions. In bioassay, 65-85% of L2 from an insecticide-susceptible laboratory strain died when fed potato foliage treated with novaluron at 25 and 50 g a.i./ha, respectively, 28 days after application. Thus, the selection pressure for novaluron-resistant CPB may be intense long after population densities have been reduced below an economic threshold level. Studies were also conducted to identify geographic variability and potentially problematic CPB populations before the widespread use of novaluron in the field. The susceptibility of L2 to novaluron was determined for 27 different populations collected from 6 Canadian provinces in summer of 2003. Despite no previous exposure to novaluron, the populations differed considerably in susceptibility with mortalities at a diagnostic dose ranging from 55-100%. Our results suggest that despite the potential of novaluron in CPB management, the possibility of rapid development of resistance exists.

Bemisia tabaci, **biotypes and insecticide resistance management.**

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Many insect species exist as a complex of biotypes that can be differentiated by a variety of genotypic or phenotypic parameters. Some of these are of ound importance in pest management. For example some biotypes of the cotton whitefly (*Bemisia tabaci*) appear to be associated with different ecological, behavioural and insecticide-resistant characteristics. The processes that mediate the balance and evolution of such co-existing forms are neither well-described nor understood. It is clear however, that in some instances, the evolution of biotypes among pest species is influenced by the tools used for their control (e.g insecticides). In an effort to understand the factors that influence the dynamics of co-existing biotypes and how these impact upon the implementation of management strategies and the dynamics of insecticide resistance, we examine Mediterranean populations of *Bemisia tabaci*. We present work that characterises insecticide resistance in recently collected strains of 'B' and 'Q' biotypes, and examine the ability of these forms to introgress (by observing courtship behaviour, mating success, hybrid sex ratio and hybrid fecundity). We examine the consequences of such introgression for the inheritance of insecticide resistance.

Hydolysis of organophosphorus and pyrethroid insecticides by insect carboxylesterases modified by *in vitro* mutagenesis

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Resistance of *Lucilia cuprina* to Ops is due to mutations in the gene encoding carboxylesterase E3 that enhance its ability to hydrolyse insecticides. Mutagenesis *in vitro* and expression of these modifications (G137D and W251L) have confirmed their role. These and nearby amino acids were substituted by others expected to affect the efficiency of the enzymes in hydolysing OPs and pyrethroids. Various substitutions of W251 had a broadly similar effect to W251L on OP hydolysis, but some enhanced malathion carboxylesterase. With the exception of F309L, nearby mutations generally diminished OP hydolysis. W251L was the most effective substitution to improve permethrin hydolysis (up to 130-fold) whilst decreasing the trans:cis hydolysis ratio to favour the more insecticidal isomer Other mutations around the catalytic site generally enhanced pyrethroid hydolysis relative to the wild type, the most effective being F309L, which enhanced transpermethrin hydolysis more than W251L. Complementary assays with individual enantiomers of the dibromo analogue of cispermethrin showed the wild type and most mutants to have a marked preference for the least insecticidal 1S conformation of the cyclopropanecarboxylate, but this was reversed in mutants with the F309L substitution. The results show the potential to design such recombinant enzymes for bioremediation of insecticide residues.

Enhancing biodiversity in sugar beet through appropriate management of GMHT crops

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The UK Farm Scale Evaluations of the environmental impact of genetically modified herbicide-tolerant (GMHT) crops reported that the biodiversity of wildlife in GMHT beet was adversely affected compared to that in the conventional crops. This was attributed to better control of weeds by glyphosate, to which the GM beet was tolerant, with knock-on consequences for the invertebrates that depended on those weeds for survival. In other separate studies on sugar beet, alternative glyphosate programmes, involving band sprays and/or delayed sprays (compared to the aft label recommendations) resulted in much weedier plots than under conventional regimes at different times in the season, with consequent benefits for some invertebrates. In many cases this was achieved with no loss of yield compared to conventional systems. Another regime that relied on one early spray of glyphosate resulted in greater numbers of late germinating weeds than in conventional sytems, and consequently greater seed production by those weeds for the benefit of seed-eating beetles and autumn-feeding birds. GMHT technology with regard to beet is so flexible that almost any scenario can be created and still allow farmers to grow a itable crop.

Ecdysteroid agonist insecticides – mechanism, selectivity and importance in IPM

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Non-steroidal ecdysone agonists belonging to the bisacylhydrazine class of compounds are a new generation of insecticidal compounds that cause premature lethal molts in susceptible intoxicated insects. Of these, three bisacylhydrazine compounds, tebufenozide (MIMIC®, CONFIRM®, ROMDAN®), methoxyfenozide (INTREPID®, RUNNER®, PRODIGY®, FALCON®) and halofenozide (MACH 2®) have been commercialized. Both tebufenozide and methoxyfenozide are predominantly toxic to lepidopteran pests of cotton, corn and other agronomic plants. Halofenozide, a soil insecticide, is toxic to both lepidopteran and coleopteran pests. All three insecticidal compounds manifest their toxic action in susceptible insects via interaction with the ecdysone receptor, which is also the target of the insect molting hormone, 20-hydoxyecdysone. This selective toxicity of both tebufenozide and methoxyfenozide to lepidopteran pests is largely due to their high binding affinity to EcRs from those insects. The toxic action of halofenozide, which is a weak binder of EcR, is primarily due to its relatively higher metabolic stability in susceptible insects. The mode of action of the three insecticides at the whole insect, ultra-structural and biochemical levels will be discussed and data shown to demonstrate their safety to beneficial insects, mammals and the environment. The use of these insecticides in IPM and IRM programs will be emphasized.

Prodigy™ - a new IPM-compatible product for insect control in Australian crops

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Methoxyfenozide (formulated as Prodigy™ 240 SC and previously coded as RH 2485), is a moult-accelerating compound (or MAC), from the same chemical family as tebufenozide (Mimic™ 700 WP), a product which has been in the Australian market for many years. Methoxyfenozide is generally active at lower rates and has a wider spectrum of activity than tebufenozide, even though activity of both is strictly limited to the Order Lepidoptera. This selectivity makes methoxyfenozide an ideal candidate for use in integrated pest management (IPM) programmes as it has no direct effect on non-lepidopteran predators and parasitoids. As the only MAC compound used in Australia against helicoverpa, with no documented field resistance methoxyfenozide is a valuable new tool for inclusion in helicoverpa resistance management strategies. It's introduction reduces selection pressure on older products and further delays the onset of resistance. Development to date has focused on helicoverpa control in cotton and tomatoes, but methoxyfenozide does have several exciting possibilities. Globally, methoxyfenozide is already recognized as having excellent activity against *Spodoptera* spp. and may prove to be a cost-effective alternative for control of armyworms in a range of Australian crops (e.g. pastures, sugarcane, cereals and turf).
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Gene resources for insecticide discovery

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The majority of insecticides and nematicides in current use act on only a small number of target receptors. Drawing on the resource provided by complete genome sequences, we have sought to identify and validate novel targets for nematicides and insecticides. The excitatory glutamate receptors involved in signalling at the insect neuromuscular junction are an attractive, but poorly understood candidate target. We have identified new members of this gene family from whitefly *Bemisia tabaci* and the bollworm *Helicoverpa armigera*. The availability of these gene sequences provided new insight into the diversity of excitatory glutamate receptor sub-types in insects. Pharmacological and molecular genetic approaches were used to identify novel serotonergic (5-HT$_3$) receptors regulating pharyngeal pumping in the free living nematode *Caenorhabditis elegans*. Lead compounds were assessed for their toxicity to *C. elegans*, *Myzus persicae*, *H. armigera* and *Heligosigmoides polygyrus*. A number of *C. elegans* cDNA sequences, one or more of which are believed to encode subunit(s) of the nematode's pharyngeal 5-HT$_3$ receptor were isolated. Preliminary gene knockdown data indicates that one of the cloned receptor subunits is vital for the proper development and normal pumping of the pharynx. Our data provide the first evidence for the existence of excitatory, 5-HT$_3$-like, receptors in nematodes and the first indication that receptors of this type are valid targets for pesticides.

Molecular mechanisms of phosphine toxicity and the genetics of resistance

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Phosphine (PH_3) is the only globally significant fumigant available to protect stored grain from insect infestation. Phosphine is a somewhat hydophobic reducing compound that can complex with biologically significant metal ions, partially disrupt mitochondrial function and induce cytotoxic levels of oxidative stress. Despite a complex array of potential modes of action, strong resistance (600X) to this compound in the pest insect, *Rhyzopertha dominica*, is achieved with resistance alleles at only two loci, neither of which negatively affects fitness. Phosphine resistant mutants generated in the model organism, *C. elegans*, have an unusually long lifespan. One of these mutants is completely resistant to the toxic effect of hyperoxia in conjunction with an otherwise sublethal exposure to phosphine. Mutant and wild type nematodes exhibit no differences in terms of phosphine induced changes to either mitochondrial morphology or ATP levels. The mutant, however, appears to be protected from phosphine induced oxidative damage to lipids. Surprisingly, genome-wide microarray analysis in *C. elegans* does not implicate mitochondrial dysfunction or oxidative stress as the primary causes of phosphine toxicity. Rather, the stress response appears to mimic the expression profiles associated with the lifespan extension mutant, *daf-2*, as well as the unfolded protein response induced by denatured proteins.

Pyrethroid insecticide resistance and treated bednets efficacy in malaria control

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Metabolic and k resistance mechanisms were characterized in An. stephensi and this easily colonized species was used to evaluate the efficacy of ITNs. Bioassays performed on 4th instar larvae showed a resistance ratio of 182. The resistance was partially suppressed by PBO (54%) and DEF (51%) or both (68%). Biochemical assays revealed that esterases, monooxigenases and glutathione S-transferase levels were 1.72, 1.86 and 1.88-fold respectively in the DUB-S compared to the susceptible strain Beech. The overall permethrin metabolism rates by DUB-S mosquitoes measured by HPLC were 1.5-fold more than by Beech strain. The classic k mutation was detected in II6 of the para-type sodium channel gene from DUB-S strain. Bioassays performed on the resistant DUB-S mosquitoes using different pyrethroid-treated bednets showed that except for deltamethrin and a-cypermethrin treated bednets, the rest of the tested pyrethroid-treated bednets at standard WHO recommended doses did not produce adequate mortality rates. The deterrency test performed on susceptible and resistant An. stephensi showed that there are significant differences between the entry rates of susceptible and resistant mosquitoes into the exposure tube containing permethrin treated bednet. These data show that when mosquitoes have both k-type and metabolic resistance mechanisms, the efficacy of pyrethroid-treated bednets is questionable.

Update on the honey bee genome project

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Along with their longstanding importance for agriculture, honey bees (*Apis mellifera*) have been important models for behavior, physiology, ecology, and genetics. They play a considerable role in evolutionary comparisons across insects and in attempts to understand the forces that lead to, maintain, and challenge insect societies. A aft sequence for the honey bee genome was funded in 2002 by the U.S. National Institutes of Health, and Department of Agriculture, following a 'white paper' competition to identify compelling eukaryotic sequencing targets. The project began that year at the Baylor College of Medicine Human Genome Sequencing Center (www.hgsc.bcm.tmc.edu), with input from individual scientists and the Sanger/Wellcome Trust. The current assembly (version 1.1, January 2004) reflects 1.25 Gbp of mainly short-insert (shotgun) sequences, giving an estimated 6.0-fold coverage of the 210 Mbp genome. The assembly is comprised of 11,600 scaffolds (N_{50} = 223 kbp) containing 30,074 contigs (N_{50} = 19 kbp). Approximately 95% of known EST's, cDNA's and markers are in this assembly. Current plans are to incorporate reads from an AT-biased library and an existing BAC library and to generate cDNA's useful for gene calling. An updated assembly and an initial gene list are anticipated by the time of this meeting.

EST analysis of genes expressed in the different tissues of the spruce budworm, *Choristoneura fumiferana*

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More than 35,000 sequence reads have been generated from five cDNA libraries of the epidermis, midgut, fat body, whole larvae and cell line of the spruce budworm, *Choristoneura fumiferana*. These expressed sequence tags (ESTs) have been assembled into approximately 8000 unigenes using Phrap program. Annotation of these sequences has been conducted against the protein database Swiss-Prot/TrEMBL and the GenBank of NCBI. The unique sequences have been classified for their molecular function, cellular location, and biological process using the Gene Ontology classification system. A spruce budworm EST database has been established and is available to the scientific community. These annotated EST sequences will serve as a resource base for gene mining, comparative analysis and genome mapping. The ESTs have also being used for developing cDNA microarrays, which will be used to study the global gene expression in the spruce budworm in response to different stresses. This project is supported by funds of Genome Canada and Canadian Biotechnology Strategy.

Implementation and impact of transgenic Bt cottons in Australia

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Bt cottons (INGARD®) expressing the CryIAc endotoxin, were commercialised in Australia in 1996/97 through an industry agreed deployment strategy which limits use to 30% of total cotton. A comprehensive management strategy minimises the risk of Bt resistance in *Helicoverpa armigera*, the target pest. In 6 years of commercial use Bt cottons have reduced pesticide applications for *Helicoverpa* by an average of 50%, providing major environmental benefits. Fibre quality has not been affected, while in some cases yields have increased. Despite early difficulties with variable performance, growers have rapidly adopted the technology and will gain long-term benefits through enhanced environmental sustainability, as Bt cotton is used in environmentally sensitive areas. Perhaps more importantly Bt cottons show great potential for IPM systems where a combination of soft approaches yields considerable benefits. That single gene Bt cotton has not been a "magic bullet" is fortunate in demonstrating that Bt cottons need to be introduced within an IPM system and supported by rigorous resistance management strategies. Two-gene Bt cottons expressing Cry 1Ac and Cry 2Ab (Bollgard II) have now been commercialised and will replace INGARD® in 2004/5. BGII provides enhanced efficacy and further reductions in pesticide requirement with associated environmental benefits. BGII also bring reduced resistance risk, but will remain part of a conservative resistance management strategy similar to that for INGARD varieties.

Pleiotropic effects of insecticide resistance genes in an aphid pest on vulnerability to attack by parasitoid wasps

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Studying the phenomenon of insecticide resistance provides a rare and valuable insight into the fundamental processes underlying evolution as resistance often appears over short timescales in human terms. In effect, this process represents a classic case of 'forced evolution' and as a result can produce resistance mechanisms that have negative pleiotropic effects on fitness in the absence of insecticides. The paper describes experiments done at different spatial scales measuring the vulnerability of insecticide-susceptible and –resistant peach-potato aphids (*Myzus persicae*) to attack by the parasitoid, *Diaeretiella rapae*. It also discusses the potential impact of the findings on Integrated Pest Management strategies utilising beneficial insects.

Insect AChE

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Two genes encode acetylcholinestesterase (AChE) in insects, but only one seems to be expressed in the synapse of the central nervous system. Protein is a dimer anchored to the presynaptic memebrane via a glycolipid. Structure has been solved for the *Drosophila* enzyme, the active site is buried 20 Å inside the protein, at the bottom of a narrow gorge. This structure results to complex kinetics resulting from the traffic of substrate and products in the active site gorge. Another consequence of this structure is the sensitivity to insecticide: insect AChE is more sensitive to insecticide than its vertebrate counterpart. This property has been used to design biosensors to detect insecticide residues and the enzyme has been engineered to improve its capacity of detection. Extensive use of insecticides resulted in insecticide resistance. Several mutations have been evidenced. They often combined in the same site resulting in an enzyme more resistant and with a broader spectrum of resistance. Mutations affected enzyme efficiency : they decrease or increase the limitating step of substrate hydolysis, deacetylation and decreases enzyme stability. This alteration of enzyme has been compensated in most of strains by an overproduction of the enzyme.

First season of methoprene application mirroring Fire Ant Control Centre treatment program has not had any prompt deleterious effects on the native soil invertebrate assemblage.

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In response to the accidental introduction of the invasive ant *Solenopsis invicta* (Buren) to Brisbane, Australia, an eradication program was mounted. This program employs a range of chemicals including the insect growth regulators methoprene and pyriproxifen, which though specific to invertebrates and intended solely for *S. invicta*, may have deleterious effects on other invertebrates. The impact of Methoprene on soil invertebrates and nutrient cycling is being evaluated using a control-impact study conducted in eucalyptus woodlands where *S. invicta* is absent. Methoprene is applied to treatment sites following Fire Ant Control Centre procedure, with 3 applications of 2 kg/ha each treatment season (November to April). Soil samples are collected: before treatment; and one month after each subsequent treatment over two treatment seasons (Nov 2002-May 2004). Soil invertebrates are extracted and sorted to morphospecies to form a community dataset for each site. Due to their numerical dominance, and also importance for processes such as decomposition and nutrient cycling, only the mite component of the invertebrate community will be reported here. Results at the completion of the first treatment season indicate that the application of Methoprene is not associated with a decrease in any of the diversity parameters. Community data are also analysed using ordination techniques to investigate whether the actual species composition is influenced by methoprene application.

Large-insert BAC libraries for fundamental studies in Lepidoptera

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The tobacco hornworm, *Manduca sexta*, the tobacco budworm. *Heliothis virescens*, and the Müllerian mimic, *Heliconius erato*, are well-established lepidopteran models for studying fundamental problems in olfaction, neurobiology, and development (*M. sexta*), evolution of insecticide resistance and plant host specialization (*H. virescens*), and wing pattern mimicry (*H. erato*). We have constructed two separate BAC libraries for each species using *Bam* HI and *Eco* RI partial digestion. Average insert sizes of *Bam* HI and *Eco* RI libraries are 152 kb and 165 kb for *M. sexta*, 171 kb and 150 kb for *H. virescens*, and 175 kb and 153 kb for *H. erato*, with <5% non-recombinant clones. Genome coverage totals an estimated 12.9x for *M. sexta*, 16.3x for *H. virescens* and 15.4x for *H. erato*. We are screening the libraries with well-conserved single-copy genes to better estimate genome coverage and obtain BAC clones to use as anchor loci for comparative studies. All libraries are publicly available at http://hbz.tamu.edu. We are indebted to L. Riddiford, F. Gould, and O. McMillan for insects, and M. Cornwall and N. Pierce for vouchering specimens. Supported in part by NSF grant no. IBN-0208388 to MRG, HBZ, and CW.

Functional genomics of the midgut of the cotton bollworm, *Helicoverpa armigera*

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The Lepidopteran insect, *Helicoverpa armigera,* or cotton bollworm, is a major pest of agriculture worldwide. Many control strategies involve orally-transmitted control agents that target the larval midgut. A functional genomics study of this organ has the potential to increase understanding of its molecular and cellular biology and to identify new targets for pest control. We have undertaken an EST project to identify genes important for midgut function, growth and development and gene/enzyme pathways involved in insect-plant and insect-pathogen interactions. Sequence analysis of midgut cDNA libraries now covers ~5,000 ESTs each from young (2nd and early 3rd instar) and from 5th instar larvae. Clustering yielded a unigene set of ~3,900 contigs. Searches against the UniProt (EBI) database and model organism proteomes yielded confident Gene Ontology annotation for ~55% of the contigs. These include genes associated with key midgut functions: digestive hydolases; nutrient and ion transport and pH maintenance; a range of detoxification enzymes and genes forming the peritrophic membrane or involved in midgut cell proliferation, differentiation and adhesion. Many have not previously been identified in lepidopteran systems. Additionally, comparison with silkworm ESTs identified likely orphan lepidopteran genes. Microarrays based on the non-redundant gene set are being established for expression iling studies.

Pesticide resistance in Brazil and IRAC-BR activities: current situation and perspectives

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Recognition of pesticide resistance problems in Brazil dates of the late 1960s, but it was not followed by further studies until the 1990s, when the interest was renewed due to reports of insect-pest control failures. The establishment of the Brazilian Insecticide Resistance Action Committee (IRAC-BR) in 1997 was a reflex of this concern and provided new impetus to pesticide resistance studies in the country. The major activities of the IRAC-BR have been centered on institutional, research and educational programs. The institutional program is based on the promotion of the Committee and its activities, while the research program establishes priorities and provides support for projects of interest. At present, the research effort is centered in two long-standing projects, *Tuta absoluta* in tomatoes and *Spodoptera frugiperda* in maize/cotton, and several other shorter-term projects. The educational program provides basic training in Insecticide Resistance to a diverse public. Annually, IRAC-BR offers short-courses, workshops, lectures and round-table discussions in different regions of the country. IRAC-BR also sponsors a National Research Award, in two categories, undergraduate and graduate research projects. This award was created to stimulate creativity, positive criticism and human resource formation in Insecticide Resistance Management in Brazil. These initiatives from IRAC-BR have been well received in Brazil and the fruits from this effort are beginning to ripen.

Esterases mediate resistance to biological insecticides in *Helicoverpa armigera*

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The cotton bolloworm *Helicoverpa armigera* is a serious pest of cotton and other summer crops in Australia, where it has a long history of insecticide resistance to most insecticides. Particularly critical for Australian agriculture was the evolution of pyrethroid resistance. Pyrethroid resistance which was found to be largely due to overproduction of specific esterase isoenzymes (R_m 0.24 - 0.33), which sequester and hydolyse pyrethroids. Modern pest resistance management strategies have incorporated new biological insecticides with novel modes of action. Such biological insecticides are spinosad and *Bacillus thurlingiensis* toxins and as well insecticidal proteins of B thurlingiensis which have been incorporated into genetically engineered plants. Nonetheless, there is an enduring threat of resistance. *H. armigera* has already developed resistance to spinosad and some CRY toxins and in this paper we report esterase mediated resistance mechanisms.

A microsomal esterase confers insecticide resistance in *Spodoptera littoralis*

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Spodoptera littoralis (Lepidoptera: Noctuidae) (Boisduval), the Egyptian cotton leafworm, is a major pest of cotton and vegetable crops in Mediterranean Europe and Africa. A field strain collected from cotton at Adana in Turkey was found to have a resistance factor of 8-fold against chlorpyrifos by leaf dip bioassay. Preliminary studies of AChE sensitivity against chlorpyrifos-oxon indicated no evidence of insensitive AChE. An enhanced microsomal esterase was revealed that correlated with the observed resistance when compared to a susceptible standard strain. We report here on the purification and kinetic parameters of this microsomal esterase.

Transgenic corn and rootworm: ecology, evolution and management.

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The western corn rootworm in North America depends almost exclusively on corn for larval survival and adult nutrition. Two-year crop rotations were very effective at controlling rootworms since oviposition historically only occurred in corn. In recent years however, crop rotation has failed in the corn-belt east of the Mississippi river. Adults in this region lay eggs in fields other than corn and because of rotation these eggs hatch in corn the following year. The change in the behavior of adult rootworms resulted in the increased use of insecticides and a renewed interest in developing ecologically sound control strategies. We report on a series of modeling efforts that examined how land use lead to the evolution of behavioral changes in adults and how landscape diversity and weather patterns contribute to the dispersal and ultimate distribution of this rotation resistance variant in North America. We then examine a variety of control options combining cultural practices with transgenic crops and other crop technologies to simultaneously manage rotation resistance and the risk of developing resistance to transgenic corn. The results were sensitive to landscape diversity, toxin dose, expression of resistance, initial allele frequency and density-dependent larval survival.

EST-cDNA clones were mapped based on RFLP in the silkworm, *Bombyx mori* L.

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We have developed the genetic analysis by using molecular markers based on restriction fragment length polymorphism (RFLP) in the silkworm, *Bombyx mori*.. In the silkworm there is no crossover on female chromosomes, so a (RF02 x RF50) female was crossed to a RF02 male and from this BC_1 segregant more than 250 ug individual DNAs were prepared and used to know their genotypes. Every clone gave homozygous or heterozygous genotype for each individual and the patterns for each clone of 15 BC_1 segregants could be identified from each other. After the 28 patterns for every chromosome were made, every new clone was easily to know their linkage group from the genotyping of 15 individuals. After linkage analysis, all clones were mapped by using the 100 segregants of the cross between a (RF02 x RF50) male and a RF02 female. From all of the genotypes, non-recombinant individuals are those with either all A, homozygous, or B, heterozygous, but recombinant individuals have both A and B. The linear order of clones is known after data is sorted and distance between clones determined by counting the recombinants. Now about 300 cDNA clones have been mapped on molecular genetic map.

Molecular tools for reconstructing the past and predicting the future of insecticide resistance

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We have been using the tools of modern biotechnology to reconstruct the past and predict the future course of the evolution of metabolic resistance to organophosphorus (OP) insecticides in the higher dipterans *Lucilia cuprina* (Calliphoridae) and *Musca domestica* (Muscidae). Resistance in both species is due to mutations that convert a carboxylesterase enzyme into a catalytically inefficient but physiologically protective phosphotriesterase. There are a pair of allelic mutations in each species which give resistance to structurally distinct sets of OPs. The two mutations involve the same substitutions in the same residues of orthologous proteins in the two species. In order to reconstruct the history of the resistance we have been using PCR to clone and sequence the gene involved from preserved museum specimens over an 80 year temporal transect. This transect goes back over 20 years before the first use of OP insecticides. We find that at least one of the resistance alleles existed in natural populations before the introduction of the pesticides. In order to predict the future course of resistance we have been using the tools of directed *in vitro* evolution and rational enzyme design in attempts to improve the catalytic efficiency of the extant natural mutants. We have now been able to recover *in vitro* mutants which are catalytically superior in the degradation of OPs.

The management of cotton aphid (*Aphis gossypii* Glover) in Australian cotton

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Until recently, cotton aphid, *Aphis gossypii* Glover, has been a persistent but readily controlled secondary pest of Australian cotton. However, during the 1998-1999 cotton-season, control failures linked to resistance were reported for organophosphate and carbamate insecticides, with some populations controllable only with diafenthiuron. Since then resistance hotspots have developed annually over eastern Australia that have only been controllable with new chemistries. The reasons for this are complex and include seasonal differences in rainfall patterns affecting over wintering host availability for aphids and a reduction in insecticide use, particularly endosulfan, with the coincident introduction of transgenic cotton. In response, aphid management has been multi-faceted and based on a complex integrated pest management (IPM) approach. This includes an Integrated Resistance Management Strategy (IRMS) that has recently been altered to better manage the selective aphicide pirimicarb. This useful IPM chemical is usually quickly lost to high-level target site resistance but we aim to manage it through improved tactical product use.

Chemical control of western flower thrips in Australia

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Insecticides are the main method used by commercial growers to control Western flower thrips (WFT) *Frankliniella occidentalis*, **in Australia. However, as has occurred overseas, WFT are resistant to a number of insecticides and insecticide groups. Furthermore, the current field application rates of many insecticides used in Australia may be too low to be effective. This would increase selection for resistance, because insecticides are being applied at sub-lethal doses. To test this, we carried out a series of field trials. Crops tested included ornamentals, tomatoes, lettuce, strawberries, cucumber and capsicum. Six key chemicals (abamectin, acephate, dichlorvos, endosulfan methidathion and pyrazophos) were evaluated. All field trials were integrated with insecticide resistance monitoring to remove the confounding effects of chemical resistance. Field trials confirmed that many application rates used for WFT were too low to be effective. Resistance monitoring showed that abamectin, methiocarb, and pyrazophos are the only chemicals where resistance was not detected. Low-level resistance was detected against acephate, dimethoate, endosulfan, fipronil, methamidophos, methidathion and spinosad. Increasing levels of resistance were detected against chlorpyrifos, dichlorvos and maldison.**

The GMO guidelines project - developing the science in risk assessment of transgenic plants

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The GMO Guidelines Project (www.gmo-guidelines.info) has developed a conceptual framework for a methodology of case-specific ecological risk assessments of genetically modified plants. It consists of a generic framework that is constructed on five scientific sections and incorporates many different scientific disciplines: 1) Problem Formulation and Options Assessment (PFOA), 2. Transgene Expression and Locus Structure (TELS), 3. Biodiversity and Non-target Impacts, 4. Gene Flow and its Consequences, 5. Resistance Evolution and Management. The generic framework of the GMO-Guidelines is comprised of a risk assessment model, which is driven by systematically structured, detailed scientific questions. The model can be applied to any case and the information and data for answering each question tailors the assessment to the specific context yielding a case- and country-specific risk assessment. We applied the framework to three case examples: Bt-maize/Kenya, Bt-cotton/Brazil and Bt-cotton/Vietnam. In our presentation, we will briefly present the major outcomes of these case studies. Several unique features of the Project: a) We work collaboratively with scientists from developing countries and jointly conduct the case study risk assessments. Thereby, we build scientific capacity and build confidence in their own scientific capabilities; b) We frame risk assessment in a multi-stakeholder deliberative process of PFOA.

Modification of sucrose synthesis in GM aspens: effects on leaf-beetle preference.

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GM forest trees potentially pose ecological problems beyond those faced by transgenic agricultural crops because: forest trees are essentially undomesticated which makes escape of transgenics to the wild more probable and trees also have long life spans, dominate the forest ecosystems and define the structure of many communities. Therefore, it is of outmost importance that we gain basic knowledge of the potential ecological effects of transgenic tree species. The aim was to determine if genetic modifications of aspens, not primarily intended to influence natural enemies, will lead to changes in the plants nutritional quality and secondary chemistry, thus effecting interactions between natural enemies (insects) and GM trees. We used GM (*Populus tremula x tremuloides*) aspens modified with respect to increased sucrose-synthesis (ZEA SPS), resulting in increased mesophyll sucrose concentration, dry mass and growth. Two different lines (different degrees of up regulation) were used and unmodified wild-type aspen served as controls. We used two common leaf feeding chrysomelid beetles, *Lochmeae caprea* and *Phratora vitellinae* in cafeteria test. *Phratora vitellinae* utilized transgenic aspens marginally less than the wildtype whereas no difference was found for *Lochmeae caprea* preference. The relationship between these results and plant primary and secondary chemistry and its implication for plant-herbivore interactions is discussed.

Biorational insecticides – important components in pest management of cotton, vegetable and ornamentals, an overview

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There have been efforts to develop biorational insecticides with selective properties to act on biochemical sites present only in the target arthropods and not in non-target organisms. This approach has led to the development of insecticides such as the neonicotinoids (which affect nicotine acetylcholine receptors), the benzoylphenyl ureas (BPUs) and buprofezin (which inhibit chitin synthesis), and the juvenile hormone (JH) mimics and ecdysone agonists. Compounds originating from natural products, which act on GABA gated chloride channel, glutamate receptors and the chloride channels (such as the avermectins and the spinosyns) and at the mitochondrial level (chlorfenapyr) with much more association to the insects than mammals, have been developed and used successfully to combat diverse insect pests and spider mites. Additional new groups of insecticides include the phenylpyrazoles (fipronil), the pyridines (pymetrozine) and the oxadiazines (indoxacarb), which act on GABA receptors, on the nerves controlling the salivary pump and on the sodium channel, respectively. All the above, are considered biorational, or "low risk" insecticides, which have relatively low detrimental effect on the environment and its inhabitants, rendering them important potential components in IPM programs. The selectivity of these biorational insecticides to beneficials and resistance evolution in key pests will be discussed.

Selection of insecticide resistant strains of African malaria vector mosquitoes

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Insecticide resistance in the major African malaria vector mosquitoes seriously impacts on control operations in many countries. In 1999/2000, South Africa experienced its worst malaria epidemic in over 50 years, due to the return of pyrethroid resistant *Anopheles funestus*. The situation is confounded by cross-resistance to carbamates in *An. funestus* and low level resistance to DDT and pyrethroids in the other southern African vector *Anopheles arabiensis*. Resistant strains of both species have been established and laboratory investigations into the mechanisms of the resistance carried out. Only a metabolic mechanism appears to be responsible for the pyrethroid resistance in *An. funestus*. In *An. arabiensis*, however, two mechanisms have been found in populations from different parts of Africa, one a knockdown (*kdr*) type resistance and the other a metabolic mechanism. The possibility of cross-resistance to other classes of insecticides is discussed.

Actions of clothianidin and related neonicotinoids on recombinant and native nicotinic acetylcholine receptors

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Imidacloprid and related neonicotinoids are insecticides acting selectively on insect nicotinic acetylcholine receptors (insect nAChRs). Among neonicotinoids, imidacloprid has pyridine and imidazolidine moieties, whereas clothianidin lacks both moieties. Since it was conceivable that such structural differences could result in different actions of these two insecticides on nAChRs, we have investigated, using two-electrode voltage-clamp electrophysiology, the agonist actions of clothianidin and imidacloprid on *Drosophila* SAD-chicken b2 hybrid nAChR expressed in *Xenopus laevis* oocytes. Also, we have used whole-cell patch-clamp electrophysiology, to study clothianidin actions on native nAChRs in the TAG neurons from male adult American cockroaches as well as in the mushroom body neurons from *Drosophila melanogaster* larvae. Clothianidin was found to activate not only the recombinant SADb2 hybrid nAChR but also native insect nAChRs with a greater efficacy than that of imidacloprid. Structure-agonist action relationship analysis suggested that opening of the imidazolidine ring is likely to account for the greater agonist efficacy of clothianidin compared to that of imidacloprid.

Novaluron (Rimon), a novel IGR – cross-resistance, selectivity and importance in pest management

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Novaluron, 1-[chloro-4(1,1,2-trifluoro-methoxyethoxy)phenyl]-3-(2,6-difluorobenzoyl)urea, a novel benzoylphenyl urea acts by both ingestion and contact. It is a powerful suppressor of lepidopteran larvae such as *Spodoptera littoralis* and *Helicoverpa armigera* (by ingestion) and whiteflies such as *Bemisia tabaci* (by contact). The compound has translaminar activity thereby affecting leafminers such as *Liriomyza huidobrensis.* Its residual activity under field conditions ranges between 10 and 30 days depending on environmental conditions. Artificial rain at a rate of 40 mm/h applied 5 and 24 h after novaluron treatment in a cotton field had no appreciable effect on the potency of novaluron on *S. littoralis* larvae. Novaluron has no appreciable cross-resistance with the juvenile hormone mimic pyriproxyfen or with the neonicotinoids. Hence these compounds can be used as components in insecticide resistance management for controlling whiteflies and other insect pests in cotton and field crops. Novaluron has no appreciable effect on parasitoids and phytoseiid mites, and probably has a mild effect on other natural enemies. Hence, it can be considered an important component in IPM programs.

The susceptibility of *Aedes aegypti* from northern Queensland to synthetic pyrethroids

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Aedes aegypti is the only mosquito vector of the dengue virus in northern Queensland. Despite complete reliance upon synthetic pyrethroids to control adult *Ae. aegypti* during dengue outbreaks, little is known regarding the susceptibility of *Ae. aegypti* from northern Queensland to these insecticides. Insecticide exposure bottle assays and biochemical microplate assays were conducted to examine the susceptibility of *Ae. aegypti* to synthetic pyrethroids. The susceptibility of five strains of *Ae. aegypti* from northern Queensland to five commonly used synthetic pyrethroids was compared with that of an insecticide-susceptible laboratory strain. All strains tested were fully susceptible to the synthetic pyrethroids used by mosquito control authorities in northern Queensland (lambda-cyhalothrin and deltamethrin). However, some wild strains showed significantly reduced susceptibility to permethrin, bioresmethrin and alpha-cypermethrin. Biochemical microplate assays were used to measure the activity levels of two types of enzymes (β-esterases and monoxygenases) that often confer resistance to synthetic pyrethroids. Elevated esterase activity was detected in three wild strains relative to the reference strain, but elevated oxidase activity was not detected in any of the mosquito strains. The presence of resistant traits in wild populations of *Ae. aegypti* demonstrated here supports the need to establish a resistance monitoring programme in northern Queensland to ensure the continued success of dengue outbreak control.

Identification and sequence analysis of the chromosomal region containing *nsd-2*, a densovirus-resistance gene in *Bombyx mori*

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Bombyx mori densovirus (BmDNV) multiplies in the columnar cell nuclei of the mid gut epithelia in the silkworm, *Bombyx mori*. It is classified into 2 types, DNV-1 and DNV-2. Some silkworm strains were identified as resistant against DNV-1 and/or DNV-2. So far four non-susceptibility genes have been reported, namely; *nsd-1*, *Nid-1*, *nsd-2* and *nsd-Z*. However, none of them have yet been isolated as responsible genes. Studies on these genes are useful in understanding the mechanism of non-susceptibility in relation to the viral invasion and multiplication. We identified four closely linked cDNA markers with *Nid-1* and *nsd-2*. By taking advantage of *Bombyx* genome information, the BAC contigs covering these cDNA markers were expanded by screening of the BAC library with BAC end sequences, fingerprinting and shotgun sequencing of BAC clones. At the same time, backcrossed F_1 segregants were selected by virus nonsusceptibility and the linked boundary of the chromosome was defined by sequence analysis.

The use of surface plasmon resonance in characterisation of metabolic resistance

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Insecticide resistance can be the result of target-site insensitivity, metabolic resistance, or both. In general, target-site insensitivity can be readily determined by direct assays on the receptor or target involved. Metabolic resistance can often be harder to characterise, often being implicated indirectly by synergism studies or the use of model substrates. These may or may not accurately reflect the 'true' interaction between pesticide and the degradative enzyme. The use of BIAcore surface plasmon resonance technology allowed the direct interaction of insecticides with a purified esterase to be measured. Additionally such interactions are measured in real time, allowing a unique insight into the binding between resistance-associated esterase and xenobiotics.

Bt-maize and *Trichogramma* wasp – two compatible strategies in control of European corn borer (*Ostrinia nubilalis*)

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The efficacy of *Bt* maize and Trichogramma wasp (Tw) against *O. nubilalis* was tested in two field trials in 2002 and 2003. The hybrids of maize MON 810 (Bt-maize), Monumental (isolinie) and Raissa (local variety) were used. Tw was applied as preparation Trichocarp. Plant injury before harvest on untreated plot ranged from 0.75 to 2.5 per plant in 2002, and 0.22 – 1.4 in 2003. Biological efficacy of *Bt* maize was 100%, the biological efficacy of Tw ranged from 35 to 69%. The increase of yield on *Bt* maize plot reached 21 - 27% in 2002, and 7 – 15% in 2003 and on plots treated by Tw ranged from 0.6 to 17 in both years. The content of four mycotoxins in grain of maize ascertained by ELISA will be presented. Mathematical model of population dynamics of *O. nubilalis* in strips of maize treated by Tw in comparison with untreated strips will be presented. Tw can be applied in insect resistant management (IRM) of European corn borer. Strips of non-transgenic maize hybrid protected by Tw can be used as a refugee to decrease spread of Bt-resistance. The work was funded by the Ministry of Agriculture of the CR project no. QD 1360.

Fluorescence PCR techniques for detecting mutations in the sodium channel gene and for analysis of family 6 oxidase genes conferring pyrethroid resistance in *Aedes aegypti*

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Multiple mutation sites in transmembrane segment 6 of domain II of the sodium channel gene confer pyrethroid resistance in *Aedes aegypti*. Existing strategies for identification of genotypes using PCR are often difficult to use with routine success, especially in the case of heterozygotes. We sought to develop more robust PCR methods based on fluorometric PCR and primer mismatch PCR that remove much ambiguity in scoring results. Here we describe several approaches to making the scoring of resistance genotypes a more straightforward and reproducible procedure. Data are shown for *Aedes aegypti* populations from several continents expressing varying degrees of pyrethroid resistance, as well as for the two mutations known from *Anopheles gambiae*. Elevated expression of P450 oxidases can also confer pyrethroid resistance in *Aedes aegypti* populations. A conserved repeat motif in the downstream P450 reductase binding site of *Aedes aegypti* family 6 P450 genes was used as a probe for quantitative analysis of oxidase copy number and expression. Oligonucleotides were selected for conserved regions of sequence (across the *Culicidae*) and for specific oxidase sequence variants. Through fluorescence PCR analysis, this allowed a comparison of oxidase copy number and expression as well as comparison to homologous sequences in other *Aedes*, *Culex*, and *Anopheles* species.

Biokinetics in insecticide discovery

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Biokinetics is the study of uptake, distribution, metabolism and excretion of xenobiotics and is an important factor to address in the development of new chemical insecticides. Methods have been developed within Syngenta to address these factors on whole insect systems, and the plants upon which insects feed, to gain an insight into biokinetic processes. An understanding of these factors allows iterative cycles of chemical synthesis to improve biokinetic properties which ultimately lead to increased biological potency. Furthermore, biokinetics has played a major role in understanding the translation of in vitro biochemical signals to in vivo biological potency. Conversely this can occur the other way round when a propesticidal compound is tested and again biokinetics can be used to ascertain the conversion of the parent to the active primary metabolite. This was recently exemplified in cyanotropane insecticidal chemistry which demonstrated potent biological activity on sucking pests (hemiptera) but had very weak micromolar activity on the insect nicotinic acetylcholine receptor (nAChR). Biokinetics proved the rapid conversion of the parent to a stable metabolite in insects which had high nanomolar affinity to the insect nAChR thus linking the in vivo to in vitro potency of this chemical series.

Genetic variation and phenotypic plasticity in *Leptinotarsa decemlineata* under different selection pressures

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Invasive pest species provide us an opportunity to study genetic variation linked to selection pressures in natural populations. As pests are invading new areas they face new temperature regimes as well as human induced selection by pesticides. Different countries use also different varieties of cultivated crop species, which may retard insect performance. All these selection pressures are expected to shape the life history traits of a species. Finland is still free of the Colorado potato beetle *Leptinotarsa decemlineata* but on the boarder of its range. To assess beetles potential to invade Finland we compared the life-history traits of four European populations (Italy, Poland, Estonia, Northern Russia). Adult size varied very little between populations when reared on two different temperature regimes. However, developmental times differed among populations suggesting that there may be enough genetic variability for the beetle to adapt further to cooler climates. The frequency of alleles conferring resistance to permethrin was highest in Polish, and declined for Estonian and Russian populations, which suggests that the Polish population has experienced highest pesticide selection. However, resistance to aziphos-methyls was in overall high and similar in all populations. Finally, host plant variety affected little the adult size and the developmental time.

Insecticide choice in alfalfa hay may protect water quality

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Some insecticides used for controlling Egyptian alfalfa weevil, *Hypera brunneipennis,* have been detected in California's surface waters and are of concern due to their impact on water quality and toxicity to some aquatic life. To assess the impact of insecticide choice on water quality, we collected tail-water samples from on-farm alfalfa sites in the northern Sacramento Valley, CA over a 3-year period. Samples were collected during irrigation after organophosphate and pyrethroid sprays were applied. We found significant differences between insecticide classes in the mortality of *Ceriodaphnia dubia* (water flea) a test organism used to detect pesticides in water. Nearly all sites where organophosphate insecticides were used resulted in 100% water flea mortality in a 24-hout test of tail-water samples. The pyrethroid-treated sections of the same fields exhibited insignificant water flea mortality nor were any pyrethroids found to a detection limit of 50 ppt. The pyrethroids used in this study provided significantly better control of Egyptian alfalfa weevil than the organophosphates with no significant differences in beneficial insect counts. Although water runoff does not always occur in alfalfa fields, insecticide choice may be an important tool for protecting water quality in this crop.

Resistance traits of genetically modified silver birch (*Betula pendula*) against insects

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Genetic modification has recently been applied to forest trees, aiming to improve productivity, quality and resistance to insect pests and diseases. We introduced a sugar beet chitinase gene into silver birch, using the *Agrobacterium* mediated gene transfer. Chitinases have been characterised as defense related proteins, particularly in plant/fungus interactions. Fifteen transgenic birch lines showing varying levels of sugar beet chitinase 4 expression and a wild type birch were applied in a field test. Birch seedlings were planted in an agricultural field according to a randomized block desing in 2000. We conducted an inventory of spatial and temporal distribution of insect species and damages caused by herbivorous insects on birch seedlings. Systematic sampling of transgenic and wild type trees was carried out early, middle and late in the growing season 2003 using four randomized field trial blocks. Furthermore, we studied the effect of the quality of partially defoliated birch foliage on performance of *Orgyia antigua* (Lymantridae) larvae in the laboratory. Transgenic and wild type trees were artificially defoliated in nine field trial blocks. This study has been a part of a risk assessment project aiming to provide new information on the biological interactions between genetically modified trees and other organisms.

Effects of neem (*Azadirachta indica* A. Juss) on rice bug, *Leptocorisa chinensis* (Dallas)

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Ovicidal, ovipositional, nymphicidal effects of neem (*Azadirachta indica* A. Juss) on rice bug, *Leptocorisa chinensis* (Dallas), were tested under laboratory and caging conditions. Neem oil alone and azadirachtin (Aza) at 60-180 ppm did not show ovicidal effect on D7. However, a high mortality was observed on D10 in treatment with the mixture of neem oil and abamectin at a low rate and moderate mortalities in treatments of Aza at high rates > 180 ppm. Neem extracts did not significantly affect the survival of adults and the damage to rice when used as ovipositional deterrents. Treatment of 1-3 instars showed that the mixture of neem oil and abamectin achieved the best results in increasing the mortality and delaying the nymphal development, followed by treatments of Aza in a rate-dependent manner. Toxicity of Aza to various instars was also nymphal-age and observation-time dependent. No second instars in rice survived to three weeks when two sprays of Aza at 60 ppm were applied. Two sprays of Aza at 60 ppm and 1.4% neem oil significantly increased rice yield by 23-44%. Egg number was moderately reduced on Aza treated plants. Neem may be integrated into an existing IPM package for *L. chinensis*.

Overview on environmental impact of Bt-corn (Event Bt 176) on non target organisms

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Field studies were conducted in Italy to assess the potential impact of transgenic *Bacillus thuringiensis* corn on non target organisms. Bt corn was modified to express the Cry1Ab toxin against the lepidopteran pest *Ostrinia nubilalis.* We monitored above-ground herbivores, predators, and parasitoids as well as soil-related organisms such as Carabidae and Nematoda. Community structures and degree of biodiversity were compared between transgenic and isogenic crops. No significant impact of Bt-corn on plant-dwelling organisms was observed. However, remarkable differences were found in the case of *Lydella thompsoni*, a parasitoid of *O. nubilalis.* Contrasting results were detected for soil organisms. The Carabidae assemblages were similar, and any differences appeared to have no correlation with Bt maize. The biodiversity of the Nematoda was quite low for both types of crops and no significant differences were found for the majority of the cases. However, Nematoda assemblages were significantly different in Bt than isogenic crops planted in clay-rich soils. These results indicate that we should not neglect taxa of agro-ecosystems before releasing transgenic crops or any other xenobiotics.

Increased efficacy of conventional insecticides against greenhouse pests by the use of piperonyl butoxide (PBO).

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The Tobacco Whitefly, *Bemisia tabaci* (Gennadius) and Western Flower Thrip, *Frankliniella occidentalis* (Pergande) are serious insect pests of agricultural and horticultural crops throughout the world. They can cause damage through both direct feeding and also through transmission of virus. The primary strategy for control for both species remains chemical sprays. However, both have the potential for fast development of resistance due to short generation time, high fecundity, and a haplodiploid breeding system. As a result, high resistance has evolved to conventional insecticides. Results of *in vitro* assays indicated efficacious use of piperonyl butoxide could substantially reduce or nullify the metabolic resistance mechanisms conferring resistance in these species. Subsequent field trials where a pyrethroid (lambda cyhalothrin) and an Insect Growth Regulator (pyriproxyfen) were applied following a pre-treatment of piperonyl butoxide, reflected the *in vitro* findings.

Biofumigation: plant cellular disruption and water achieves high toxin levels in soil

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Some brassicas are potentially large sources of isothiocyanate (ITC) toxins, in the same chemical family as the methyl isothiocyanate produced by the widely used broad-spectrum pseudo-fumigant pesticide metham sodium. Endeavours to harness this potential in the form of biologically active green manures for IPM is dubbed 'biofumigation'. Recent laboratory studies have shown that ITC concentration in soil increased 100-fold with a combination of disruption of plant tissue at the cellular level (achieved by freezing and thawing) and high levels of moisture. Analytical determination of ITCs in soil following manipulation of *Brassica* green manure crops showed that similar improvements in the concentration of these compounds could be achieved in the field. It was crucial to thoroughly pulverise plant tissue with a club-type mulcher running at high speed. A blade mulcher tended to chop the plant tissue into fragments without liquefying it. Leaving the pulverised mulch on the surface and immediately watering it heavily achieved the greatest ITC concentration in soil. It is evident that the intrinsic water content of brassicas is insufficient to allow maximum hydrolysis of the precursor glucosinolates to ITCs. Addition of water boosts this process and carries the ITCs into the soil.

The crucial role of calcium in enhanced biodegradation of soil-applied pesticides

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Enhanced biodegradation occurs where particular, but otherwise entomologically innocuous, soil bacteria are selected by repeated use to catabolise soil-applied pesticides. It is a subtle process akin to resistance in reducing soil-borne pest control efficacy. It is more widespread than is generally recognised, possibly because of its indirect mode of action, counter-intuitive mechanism and probable confusion with resistance. Extremely severe enhanced biodegradation of the widely used broad-spectrum soil-applied pesticide metham sodium occurs in Australia in intensive horticulture, particularly on sandy soils. Highly resilient spore-forming bacteria are the cause. Pesticide persistence in affected soil was just seven hours, but 15 days in similar soil never exposed to metham sodium and in affected soil sterilised by autoclaving. A positive correlation has often been reported between the risk of onset or degree of enhanced biodegradation and soil pH. However, high agricultural soil pH is often associated with greater calcium concentration through liming (adding calcium carbonate) soils with pH below agronomically desirable levels. We have found that high soil pH alone is insufficient to exacerbate enhanced biodegradation. Rather, a powerful synergy is established when high levels of calcium occur in the same soil. Reassessment of soil pH targets and adjustment methods less reliant on calcium-rich alkalis offer scope to reduce the risk of enhanced biodegradation.

Ethanedinitrile: a promising replacement soil fumigant for methyl bromide

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Methyl bromide, a long-used powerful broad-spectrum soil fumigant, is scheduled for withawal from soil use in 2005 under the Montreal protocol. Many highly intensive horticultural industries with extreme market-iven or mandated sanitation demands face significant challenges in maintaining equivalent plant protection efficacy with alternatives. Most current alternatives have a narrower activity spectrum, greater risk of phytotoxicity entailing economic penalty in longer plant-back times, are susceptible to enhanced biodegradation (a concept alien to the methyl bromide experience) or are generally less forgiving for robust use in the highly diverse and variable field environment. CSIRO Entomology has identified and taken patent protection on ethanedinitrile (C_2N_2) (EDN) as a potential soil fumigant. Its chemical properties place it at no risk as an ozone-depletor or greenhouse gas and it is safer to use than methyl bromide. Laboratory and preliminary field tests have shown that it offers equal, if not greater, spectrum of activity and potency than methyl bromide, and an extraordinarily rapid action and rate of dissipation that promises extremely short plant-back times and little prospect that enhanced biodegradation will impede efficacy with longer-term use.

Insect resistance traits in the near future

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Since their introduction nearly 10 years ago, insect resistance (IR) traits have become a significant feature of the crop protection landscape. In spite of the rapid adoption of this technology, there are relatively few IR traits currently in the marketplace, and they are only available in two major crops (maize and cotton). A limited number of new IR traits are in the product development pipelines of the major crop protection companies with projected product launches within the next 5 years. What is known about these new IR traits will be discussed. The discovery and development process for IR traits will also be discussed in the context of identifying barriers to the introduction of new IR traits.

Efficacy of a spinosad suspension concentrate formulation for controlling chewing and sucking lice on cattle under field conditions

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Field studies were conducted with cattle in Wisconsin and Illinois between January and April, 2000, to determine the efficacy of 0.04% spray and 2 mg/kg pour-on treatments of a spinosad suspension concentrate formulation against chewing and sucking lice. Positive control substances included in the study were Co-Ral® EC (5.8% coumaphos) and CyLence™ Pour-on (1% cyfluthrin). Animals in the negative control group remained untreated. All lice species were visually sampled on each animal at weekly intervals after treatment, for eight weeks. Individual louse counts were summed over each examination site for each species. A repeated measures analysis using a mixed model (SAS PROC MIXED), was conducted for data pooled across both study sites. Both spinosad treatments and CyLence™ Pour-on provided excellent efficacy (98-100%) against *Bovicola bovis* over the entire study period; Co-Ral® EC efficacy dropped below 90% from Weeks 6-8. The efficacy of the spinosad spray treatment against *Linognathus vituli* was >93% for the entire study and >88% for the spinosad pour-on treatment. Co-Ral® EC was >90% efficacious against *L. vituli* during Weeks 1-7, while CyLence™ Pour-on only exceeded 90% for Weeks 3-7. *Solenopotes capillatus* were found only at the Wisconsin site; all treatments were highly efficacious throughout the study period.

Progress of *Bombyx* genome project in Japan

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In Lepidoptera, the genome information had been quite limited so far. Now the situation changed quickly, since National Institute of Agrobiological Sciences (NIAS) is performing the *Bombyx* genome project including *Bombyx* whole genome shotgun sequencing, cDNA collection and construction of physical map systematically in collaboration with many groups, and the explosive accumulation of *Bombyx* genome information will be expected. We will present the recent progress on our *Bombyx* genome works.
EST db: Recently, *Bombyx* EST db, “SilkBase”, was transferred to “KAIKObase” http://sgp.dna.affrc.go.jp,, where EST db is combined with genome and map information as the integrated database.
Linkage map: About 2,000 genetic markers were mapped in 28 linkage groups under the efforts of several groups. The high density map was made public as “BombMap” in KAIKObase.
BAC library: Three BAC libraries prepared by different restriction enzymes are available.
EST microarray: EST microarray containing 6,000 non-redundant ESTs was available for functional studies.
Genomic sequence: NIAS carried out *Bombyx* WGS, whose data will be public in KAIKObase of NIAS HP shortly.

Reduced sensitivity of acetylcholinesterase in green rice leafhopper, *Nephotettix Cincticeps* Uhler, resistant to organophosphorus and *N*-methyl carbamate insecticides

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Green rice leafhopper (GRLH) showed resistance to various organophosphorus and *N*-methyl carbamate insecticides. Major resistance mechanism is a reduced sensitivity of acetylcholinesterase (AChE) to these insecticides. A hydophilic form of AChE was purified from *N*-methyl carbamate susceptible (SA) and highly *N*-methyl carbamate-resistant (N3D) strains of GRLH. Both of purified AChE from SA and N3D strains displayed the highest activities towards acetylthiocholine (ATCh) at pH 8.5. In the SA strain, the optimum concentration for ATCh, propionylthiocholine (PTCh), and butyrylthiocholine (BTCh) were about $1x10^{-3}$, $2.5x10^{-3}$, and $1x10^{-3}$ M, respectively. However, in the N3D strain, substrate inhibition was not identified for ATCh, PTCh, and BTCh to $1x10^{-2}$ M. The V_{max} value in the N3D strain was 1.75- and 3.15-fold higher for ATCh and BTCh than that in the N3D strain. However, it was 1.13-fold lower for PTCh. The increased activity of AChE in the N3D strain is due to the qualitatively modified enzyme with a higher catalectic efficiency. AChE from the N3D strain was 65-fold less sensitive than SA strain to inhibition by propoxur. In contrast, AChE from the N3D strain 65-fold more sensitive to inhibition by monocrotophos than AChE from the SA strain. This indicated negatively correlated cross-insensitivity of AChE to propoxur and monocrotophos.

Large scale field trials in the USA with Bt cotton

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Field studies were conducted in 2000-2002 to compare arthropod populations between Bollgard$^{\text{Ò}}$ and conventional cotton season-long in Alabama, Georgia, and South Carolina. For each region, three or four paired fields were evaluated weekly, with each pair consisting of a Bollgard$^{\text{Ò}}$ field and a conventional cotton field (treated for lepidoptera when necessary). Insect species sampled included *Heliothis virescens*, *Helicoverpa zea*, *Spodoptera frugiperda*, *Spodoptera exigua*, *Trichoplusia ni*, *Pseudoplusia includens*, phytophagous stink bugs and plant bugs, cotton aphids, *Solenopsis invicta*, *Geocoris* spp., *Orius* spp., spiders, parasitic wasps, green and brown lacewings, and *Nabis* spp. Most results show that there were no significant differences for any arthropod population between a particular Bollgard$^{\text{Ò}}$ field and the corresponding conventional cotton field. However, the most amatic significant differences occurred in South Carolina when comparing conventional cotton sprayed with at least several applications of insecticides to control primarily *H. zea*, and Bollgard$^{\text{Ò}}$that was not treated for *H. zea*. In these instances, there were significantly more non-target arthropods in the Bollgard fields than in the conventional fields. These results demonstrate that the use of Bollgard$^{\text{Ò}}$usually has no impact on non-target arthropod populations, and when compared with cotton treated with insecticides for lepidoptera, results in significantly more non-target arthropods.

Molecular diagnosis of *para*-locus mutations in head lice (*Pediculus humanus capitis)* from around the world: their role in resistance to synthetic pyrethroids?

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Throughout the world head lice (*Pediculus humanus capitis*) infest people, and the pediculicides used to treat them do not always work. One group of chemicals applied to treat head lice are the synthetic pyrethroids, and resistance to these chemicals has been detected in lice in many parts of the world using the World Health Organisation (WHO) mortality assay. The genetic basis for resistance in lice to synthetic pyrethroids is hypothesised to be due to mutation in the *para*-orthologous sodium channel α-subunit (*para*-locus); where most of the sodium channel gene mutations that are known to be associated with knockdown resistance in insects are located. A number of point mutations have been detected in the *para*-locus of synthetic pyrethroid-resistant lice. We sequenced a region of the *para*-locus where two of these mutations have been previously found, in lice from Australia, Argentina, France, Guatemala, Thailand, Panama and the U.S. We observed the occurrence of these and other mutations and the correlation between genetic data and WHO mortality assays for many individual lice. We also developed a possible genetic test for resistance, a polymerase chain reaction restriction fragment length polymorphism (PCR-RFLP) assay to quickly and accurately diagnose individual lice as heterozygous or homozygous for genetic change to the *para*-locus.

Using genomic tools to isolate and characterise olfactory genes from the tortricid pest, *Epiphyas postvittana*

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With completion of the *Drosophila* genome, efforts have turned to insects of major economic interest for whole genome sequencing. However in the short-term whole genome sequencing will still remain beyond the reach of many projects working on individual insect species. Tools such as EST sequencing and proteomics can contribute to finding genes/proteins involved in targeted processes and create useful information for comparative genomic analysis. We are using these approaches to identify genes involved in the detection of odorants, particularly sex pheromones, from the horticultural tortricid pest, *Epiphyas postvittana*. Potential odorant binding proteins, odorant receptors and odorant hydolases have been isolated from ESTs derived from male antennae. Comparison of male and female antennal proteins on 1-D and 2-D gels reveals proteins both up and down-regulated in males relative to females, and mass spectrometry reveals some of these proteins correspond to genes from the EST collection. These techniques have given us quick access to potential olfaction genes. Functional studies of these genes to examine expression in antennae, protein structure and function, and involvement in olfaction using expression in insect cell lines and RNAi in whole insects will be described.

Expressed sequence tags from the brown planthopper, *Nilaparvata lugens* and its tissue-specific genes

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A total of about 35,000 expressed sequence tags (ESTs) were produced from various tissues and stages of *Nilaparvata lugens*, one of the serious pests of rice plant. cDNA libraries were made from two stages of eggs, fourth instar nymphs, two stages of adult ovary, testis, male midgut, female midgut, female salivary glands, female head, female thorax, female abdomen, male head and thorax, and male abdomen. There were more than 8,000 non-redundant ESTs, potentially representing unique genes. Eleven ESTs were found only from the female abdomen and ovary, and five out of the eleven were expressed only in the mature ovary. At least ten ESTs were testis-specific. Nucleotide sequences of truncated region of these cDNAs were determined by 5' and 3' RACE. BLAST search disclosed that the sequences of many gonad-specific genes were dissimilar to those in the databases. The ESTs were also used for determining whole mitochondria genome sequence, because the mitochondrial gene sequences were contaminated in the EST data. *Nilaparvata lugens* mitochondria genome size was 17,251 bp.

Prospects and potential pitfalls associated with the use of transgenic corn for fall armyworm management

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Transgenic corns that produce Bt toxins have been available for control of European corn borer in the Midwestern USA for several years. While the Bt-event in sweet corn is well expressed and also provides essentially 100% control of fall armyworms, the event in field corns provides only 60 to 85% control of this migratory, polyphagous pest. Despite the reduced potential for fall armyworm control, these transgenic products provide substantial yield gains over traditional field corns for grain and silage. With the recent development of sub-tropically adapted, Bt-enhanced field corns, acreage of Bt-corn grown for silage has spread south into the insect's overwintering region of Florida. IRM stewardship agreements standardized and enforced by the EPA to limit resistance development encourage growers to plant a proportion of their acreage to non-Bt corn reservoirs dependent upon their location within the U.S. cotton growing belts. Corn growers are increasingly pressed to produce high yields with low inputs to compete in the marketplace and are pushing to reduce or eliminate the reservoirs. These and other issues, including non-compliance and season-long availability of Bt-enhanced corn and cotton, will be examined for their potential for selection of Bt-resistance and its area wide implications for fall armyworm control.

Cost economics of Bt cotton cultivation in irrigated ecosystem

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Bollworms, *Helicoverpa armigera* (Hubner), *Earias vittella* (F.) and *Pectinophora gossypiella* (Saunders) have made cotton cultivation uneconomical. Transgenic cotton hybrids with in built δ endotoxin reduces pest problem and cost of cultivation. Studies were undertaken at Regional Agricultural Research Station (RARS) and in farmers' field. MECH-162 Bt and non Bt during 2002-03 and MECH-184 Bt and non Bt during 2003-04 and local hybrid (NHH-44) were cultivated. Sprays were imposed as and when the pest population/damage crossed ETL. Totally 3, 4 and 4 sprays during 2002-03 and 3, 12 and 12 sprays during 2003-04 were imposed on Bt, Non Bt and NHH-44 hybrids respectively in RARS, whereas, in farmers' field 4, 9, and 9 sprays during 2002-03 and 6, 12 and 12 sprays during 2003-04 were imposed on Bt, Non Bt and NHH-44 hybrids respectively. Incidence of bollworms only in Bt cotton MECH-162 and 184 was much below ETL. More good opened bolls and less bad opened bolls per plant were recorded with significantly higher seed cotton yield in Bt cotton as compared to non Bt and local hybrid. Bt cotton hybrids recorded 35 to 45 percent increase in net it over local hybrid making it commercially viable and itable.

Wound-inducible expression in transgenic plants after insect attack

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Promoters of genes that are induced by microbial pathogens, wounding and treatment with wound-signalling compound could be valuable in regulating the expression of resistance genes when plants are attacked by insects or pathogens. A plant expression vector pMP4 was developed using a GUS/GFP fusion construct driven by a *Shpx6b* peroxidase gene promoter (J. Manners, UQ) from a tropical forage legume *Stylosanthes humilis*. This construct was introduced into insect susceptible tobacco using *Agrobacterium tumefaciens*. Expression of *gus* and *gfp* reporter genes in T_1 transgenic tobacco leaves after infestation/treatment with chewing insects, sucking insects, the plant defense regulator MeJa, and mechanical wounding were investigated. *Gus* reporter gene levels were also assayed to quantify expression and the results were consistent with the observed histological patterns of expression. The results show that the *Shpx6b* promoter switches on the expression of linked genes after damage by insect herbivores, and could be useful in regulating the expression of heterologous genes for insect and /or pathogen resistance in transgenic plants.

Appraisal of cross resistance potential to neonicotinoid insecticides by *Bemisia tabaci*

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Resistance that occurs to a particular insecticide is often assumed to extend to all members of the class. However, structural differences among compounds may affect vulnerability to degradation by metabolic enzymes, or differences in target site receptors may influence susceptibility to specific insecticides, thereby mitigating cross-resistance effects. Neonicotinoids are an important class of insecticides used worldwide against *B. tabaci* and many other insect pests. Populations from southern Spain resistant to imidacloprid have also shown cross-resistance to thiamethoxam and acetamiprid. To further assess the expression of cross-resistance to neonicotinoid insecticides, *B. tabaci* populations from California and Arizona were monitored periodically to determine their susceptibilities to acetamiprid, dinotefuran, imidacloprid, and thiamethoxam. In addition, two imidacloprid-resistant strains were tested for cross-resistance potential. Field strains expressing different levels of resistance to imidacloprid did not always show cross-resistance to other compounds. For example, a population from Arizona collected in November 2003 had an LC_{50} of 115ppm to imidacloprid in a leaf-uptake bioassay, but only 8 and 9 to dinotefuran and thiamethoxam, respectively. The resistance factor was even greater in a strain from Guatemala that yielded an LC_{50} of 653ppm to imidacloprid, but only 29 to dinotefuran. Implications for cross-resistance between neonicotinoids among the various strains are examined.

Developing protocols for field evaluations of non-target organisms in Bt crops

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Transgenic crops containing insecticidal genes from Bacillus thuringiensis (Bt) bacteria offer unprecedented power to combat agricultural pests, but concerns about their possible unintended effects remain. Laboratory research suggests accidental exposure or indirect effects of Bt toxins may negatively impact non-target species, though this result is rarely seen in field studies. However, such field research sometimes employs methods inappropriate for evaluating Bt crops, which may have relatively small effects on a select number of species. Nonetheless, simple modifications to experimental protocols and analysis could greatly increase the value of field tests of non-target effects. A synthesis of existing data may guide choices of appropriate indicator taxa, needed replication, and optimal sample timing. Current statistical methods may also allow for more powerful analysis of time-series community data. Field experiments assessing the effects of plot size on community and species-level tests of non-target effects may allow for research plots which are both practical and realistic. Overall, such changes will improve researchers' power to detect differences, positive or negative, between Bt crop production and alternate management practices. However, methods developed for current Bt corn varieties should be adaptable to studies of new cultivars, non-Bt management practices, and other cropping systems.

Studies on detection and characterization of organophosphate-resistance in *Anopheles culicifacies* sibling species in Madhya Pradesh, Central India

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Field studies carried out in villages in two blocks in Chhindwara district indicated presence of malathion resistance in *Anopheles culicifacies,* the principal vector of malaria in this area, in the absence of use of malathion spraying for malaria control. *An. culicifacies* s.l. was found 36-86% resistant to malathion but was susceptible to other organophosphate, carbamate and synthetic pyrethroid insecticides. Selection of malathion-resistance in *An. culicifacies* appears to be due to insecticides used in plant protection. Biochemical assays indicated non involvement of general esterases and insensitive acetylcholinesterase in conferring malathion-resistance. Synergist-malathion bioassays indicated the involvement of specific malathion carboxylesterase (MCE). ASPCR assays have revealed prevalence of species B (73%) and species C (27%) and exhibited diferential susceptibility to malathion. The observed percent susceptibility to malathion in species B was 68% while in species C it was 13% These results will be presented in detail and the implication of development of resistance owing to selection by agricultural pesticides will be discussed.

Sugarcane borer (Lepidoptera: Crambidae) resistance management studies in Louisiana

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During the last decade, sugarcane borer (SCB), *Diatraea saccharalis* (F.) management in Louisiana has been impacted by cultural practice changes emphasizing early planting initiated in August, and the increasing dominance of a high yielding borer-susceptible variety (LCP 85-384) now approaching 88% of the planted acreage. Insecticide foliar sprays for SCB control rose from 0.2 applications per season in 1994-95 to 1.8 applications per field in 2002-03. Due to the growing popularity of the ecdysone agonist, tebufenozide (Confirm®), insecticide resistance monitoring efforts were begun throughout the sugarcane industry in 2002. Increases in LC_{50} values from traditionally low insecticide use areas have ranged up to 2.6 fold in areas averaging more than one application per season. In one area of heavy tebufenozide use, LC_{50} values remained high despite discontinuance of selection pressure. Resistance management studies will be discussed in relation to broader SCB management issues.

Twenty years of insecticide resistance management in the Australian cotton industry.

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The Australian Cotton industry has been guided by an Insecticide Resistance Management Strategy (IRMS) for twenty years. Since its introduction in the 1983/84 cotton season to combat widespread pyrethroid resistance by the primary pest of cotton *Helicoverpa armigera*, it has been modified and expanded to encompass an increased number of chemical groups, embrace integrated pest management and account for a wider range of insect pests. Compliance to this voluntary strategy is near to 100 % by the cotton industry, and in recent years the grains industry has become involved in the strategy providing a more co-ordinated attempt across multiple farming practices to manage resistance and maintain the chemicals available for pest control. This paper shall provide an overview of the IRMS, its evolution over the last twenty years, the principles involved and why uptake by the industry has been so successful. The current status of insecticide resistance by the primary pests of cotton shall be discussed, as well as the future of the IRMS, including the possible impact of transgenic cotton on the pest complex and insecticide resistance management.

Global status of insecticide resistance mechanisms in *Helicoverpa armigera*

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The cotton bollworm *Helicoverpa armigera* (Hübner) has displayed an enormous potential to resist almost all the insecticide molecules used against it thus far. Amongst the range of mechanisms that confer resistance, enhanced metabolic detoxification has been found to be the most important. Recent evidence showed that increased oxidative breakdown of pyrethroids was found to be associated with pyrethroid resistant strains from West Africa, India, China, and Pakistan. Thus far, at least four cytochrome p450 genes CYP6B2, CYP6B6, CYP6B7 and CYP4G8 have been implicated in pyrethroid resistance. Pyrethroid resistance due to enhanced esterases, reduced penetration and nerve insensitivity was reported from India, China, Pakistan, Thailand and Australia. Several genes associated with nerve-insensitive resistance were explored for specific structural changes, but the mutations generally associated with sodium channels and GABA chloride channels in other insecticide resistant species were not found in *H. armigera* and those that were reported previously in the intracellular segment linking repeat domains III and IV of the para-homologous sodium channel gene, that were associated with nerve insensitive *H. armigera* larvae, were not confirmed. Insensitive acetylcholinesterase and or unique esterase isozymes were found to be associated with resistance to organophosphates and carbamates. Resistance diagnostic kits based on the unique resistance associated isozymes, have been made in India.

Resistance issues in Bt cotton in India and China

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Genes for resistance to Cry toxins being selected in the field *in Helicoverpa armigera* by Bt cottons may not be necessarily those which are also being found in laboratory selections. Resistant individuals are present in the field at frequencies of between one in 0.42x10^{-3} and one in 1.3x10^{-3} in India and China. There appears to be both a recessive target site mechanism and a dominant enzymatic degradation mechanism. Currently the former seems likely to be more important in Chinese field populations and the latter in Indian populations with important implications for resistance management strategies. A simulation model was produced taking account of the biology of the pest, the local cropping pattern, initial resistance gene frequency, dominance, mode of inheritance, relative survival rates of RR, RS and SS individuals on Bt and non-Bt hosts throughout the year and other variables. Factors of importance in determining the time to a filed resistance gene frequency of 0.5 in the two countries are different. Where resistance is semi-dominant, in-crop refugia were of little value in delaying resistance development. However even fitness costs as low as 5% doubled the time to significant resistance in most scenarios. Integrating other artificial mortality factors at times of peak pest presence was the simplest effective strategy for delaying resistance in systems where >40% of the cotton is Bt transgenic. The likely impact on resistance development of the range of dual gene combinations currently being commercialised in the two countries has been modeled and is discussed.

Survival and development of *Spodoptera frugiperda* and two of its natural enemies on Bt transgenic maize

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This paper reports effects of conventionally-bred partially resistant and transgenic maize hybrids on *Spodoptera frugiperda* (herbivore), *Campoletis sonorensis* (parasitoid) and *Beauveria bassiana* (entomopathogenic fungus).   *Spodoptera frugiperda* larvae fed maize expressing the Bt toxin Cry1Ab had an increased development time and reduced pupal weights compared to larvae fed conventionally-bred resistant and susceptible maize hybrids. *Campoletis sonorensis* cocoon and adult weights were significantly reduced by 15-30% when reared in hosts fed Bt maize. The extent of this effect varied with the weight of host at oviposition, its subsequent development and host weight at parasitoid emergence. Resistant maize, whether transgenic or conventionally-bred, may increase the susceptibility of *S. frugiperda* to *B. bassiana*, as larvae develop more slowly than on susceptible maize, with greater time intervals between moults. This increases the time for *B. bassiana* spores to germinate and invade the host through the cuticle before they are shed during moulting. Alternatively, resistant plants may induce an immune response in *S. frugiperda*, reducing susceptibility to fungal attack. Results of ongoing experiments on this interaction will be presented.

The *Diabrotica* genetics consortium: a paradigm for global cooperation in research

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The evolution and spread of insecticide- and rotation resistance, the introduction and spread of the western corn rootworm (*Diabrotica virgifera virgifera*) in Europe, and the recent large-scale commercial deployment of rootworm-active *Bt* corn in North America have generated a growing sense of urgency among scientists involved in rootworm genetics research. These are all complex problems, with a number of researchers in several countries working on various aspects. In the last year, a group of more than 35 scientists from 25 different institutions in the U.S., Canada, France, and Germany have come together to form the *Diabrotica* Genetics Consortium. The main role of the Consortium is to facilitate communication among those working on related problems, both to avoid unnecessary overlap of effort and to reveal opportunities for synergism through cooperation/collaboration. It is founded on the principle that open communication of research goals, activities, findings, and future plans is in the best interest of all concerned and in the best interest of advancing science. The scientists comprising the Consortium are committed to sharing information and ideas in an atmosphere of mutual trust, respect, and sensitivity to the essional needs of one another in accord with the highest ideals of scientific endeavor.

Induction and transmission of Bt-tolerance in the flour moth *Ephestia kuehniella*

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The use of *Bacillus thuringiensis* endotoxins to control insect vectors of human diseases and agricultural pests is under threat from the possible evolution of resistance in major pest species. In addition to high levels of resistance, produced by receptor insensitivity, several cases of tolerance to low to medium levels of toxin have been reported in laboratory colonies of lepidopteran species. Since the molecular basis of some of these cases of tolerance to the toxin are not known, we explored alternative mechanisms. Here we present evidence that tolerance to a Bt-formulation in a laboratory colony of the flour moth *Ephestia kuehniella* can be induced by pre-exposure to a low concentration of the Bt-formulation and that the tolerance correlates with an elevated immune response. The data also indicate that both immune induction and Bt-tolerance can be transmitted to offspring by a maternal effect and their magnitudes are determined by more than one gene.

Effects of reduced prey choice caused by GM plants on a predatory insect

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Bt plants are highly toxic to certain pest species. Predatory insects may be affected by the reduced abundance of target pests if sufficient alternative prey is not available. This paper will present experiments investigating such indirect effects under laboratory conditions. Lacewing (*Chrysoperla carnea*) larvae were released into large cages containing Bt and wildtype plants previously infested with a constant number of diamondback moth (*Plutella xylostella*) eggs but different numbers of aphids (*Myzus persicae*). No *P. xylostella* larvae survived on Bt plants but aphid populations reached high densities on both plant types. Lacewings recovered from Bt plants at the end of the experiment did not differ in weight from ones recovered from wildtype plants. At the highest initial aphid densities there was also no difference in the proportion of lacewings surviving on the two plant types. Significantly fewer lacewings were recovered from Bt plants than from wildtype plants as initial aphid numbers were reduced. Interestingly, however, numbers of *C. carnea* recovered from wildtype plants were only significantly correlated with numbers of surviving *P. xylostella* at the highest aphid densities. Possible roles of escape behaviour of *P. xylostella* as well as cannibalism between *C. carnea* larvae in these interactions will be discussed.

The effect of spinosad on different developmental stages of Colorado potato beetle *Leptinotarsa decemlineata* (Say) (Col.: Chrysomelidae)

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The Colorado potato beetle (CPB) *Leptinotarsa decemlineata* is a major insect pest of potatoes. In this study we investigated the effect of spinosad on egg, larval and adult stages of CPB. A strain of CPB was collected from Ardabil, where this pest was reported for the first time from Iran in 1984; and was reared in greenhouse conditions. Bioassays were conducted using eggs, neonates, 1st, 2nd, 3rd and 4th instar larvae as well as adults. In preliminary experiments, no ovicidal effect was observed. The concentrations used for treating the larval stages and adults ranged from 1-80 ppm. To estimate the LC values, each treatment was replicated 4 times. Bioassays were conducted at 26 ± 1 °C and 16:8 (L:D) h photoperiod in transparent plastic containers. The mortalities were recorded 24 hrs after treatment. Based on probit analysis of the data, LC_{50}s were 2.1, 3.2, 4.6, 6.5, 20.2 and 12.0 ppm and the estimates for LC_{90} values were 8.0, 12.9, 17.0, 30.1, 67.8 and 30.2 ppm for neonates, 1st, 2nd, 3rd and 4th instar larvae and the adults, respectively. If spinosad also performs well in the field, this insecticide will be a suitable candidate for CPB management programs in the region.

Laboratory selection for resistance to Bt toxins in the European corn borer: potential and limitations

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Transgenic maize hybrids expressing toxins from *Bacillus thuringiensis* have been available for managing European corn borer (*Ostrinia nubilalis* Hübner) populations since 1996. However, there is concern that this technology will result in widespread resistance to Bt toxins. The "high dose-refuge strategy" is the currently accepted method for managing resistance in *O. nubilalis* and has become a requirement of registration for all transgenic events. However, this strategy is based on a number of assumptions that are difficult to validate until resistance is identified and characterized. Resistant strains resulting form laboratory selections provide a potential tool to assess these assumptions in the absence of field derived resistant populations. However, such laboratory experiments are limited by their inability to reflect the selection intensity imposed on a field population exposed to transgenic plants. Our laboratory has been selecting *O. nubilalis* by exposure to Bt toxins incorporated into an artificial diet since 1998. A number of different colonies that vary with regard to the nature of the resistance, degree of dominance, number of genes and ability to survive on transgenic plants have been developed. The relevance of these strains in terms of their ability to improve and refine resistance management recommendations will be discussed.

Insecticide mode of action classification: a key to insecticide resistance management

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The Insecticide Resistance Action Committee (IRAC) has developed a classification scheme for insecticides (and acaricides) that is based on mode of action (MoA). Experience has shown that effective insecticide resistance management (IRM) strategies seek to minimise the selection for resistance for any one type of insecticide or acaracide. In practice, alternations, sequences or rotations of compounds from different MoA groups provide a sustainable and effective approach to IRM. This ensures that selection for resistance through the use of compounds in the same MoA group is minimised. It is recognized that insecticide resistance can also result from enhanced metabolism by enzymes within the pest, reduced penetration of the pesticide into the pest or behavioural changes of the pest. These resistance mechanisms are not linked to any site of action classification, but are specific for individual compounds or chemical groupings. Despite this, alternation of compounds from different chemical classes remains an entirely viable resistance management technique because such a practice will always minimise selection pressures. This presentation describes the mode of action classification scheme and its use in IRM.

Insecticide resistance surveillance of disease vectors in Peru

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Vector-borne diseases, such as malaria and dengue, are significant public health problems in many countries of South America, including Peru. While environmental modifications and infrastructure improvements to reduce vector populations are the ideal, the reality in many areas of the world is that insecticides play a crucial role in vector suppression. Resistance to these insecticides can potentially interfere with vector control operations. In Peru, there is little information concerning insecticide resistance in *Anopheles darlingi* or *Aedes aegypti*, the principal vectors of malaria and dengue, respectively. Utilizing the CDC bottle bioassay, personnel from the U.S. Naval Medical Research Center Detachment, Lima and the Peruvian Instituto Nacional de Salud, have conducted over 3,300 assays of over 70,000 *An. darlingi* and *Ae. aegypti* mosquitoes to establish diagnostic concentrations for 8 insecticides representing 3 chemical classes. In addition to the assays, a workshop was held to train 22 technicians from 11 regional reference laboratories in Peru on the CDC bottle bioassay methodology. The goal of these assays and training is to ultimately establish a national insecticide resistance monitoring program and assist the Peruvian health authorities in implementing effective vector control programs.

Mechanisms underlying mite outbreaks following imidacloprid applications: effects on mite fecundity and natural enemy communities in landscapes.

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Imidacloprid, a chloronicotinoid insecticide has become one of the most widely used products for insect control. It provides excellent control for many pests; however, following the elimination of the primary pest, secondary infestations of mites have been observed. The mechanisms underlying this phenomenon are thought to include alterations in mite fecundity levels stimulated by sublethal levels of the insecticide (hormoligosis), and disruption of natural enemy communities. To investigate the effect of imidacloprid on beneficial insects, we examined the influence of the insecticide on longevity and survival of a general predator, *Orius insidiosus*, and abundance and diversity of natural enemies on imidacloprid treated plants. Hormoligosis in boxwood mite (*Eurytetranychus buxi*) was also investigated. *Orius insidiosus* was shown to be negatively affected by imidacloprid in plants. However, the study of insect diversity and abundance indicated that beneficial insect communities differed only slightly between imidacloprid treated and untreated sites. Moreover, we were able to create increased densities of mites and attendant damage in the absence of predators. The results of the study suggest that the mechanisms leading to increased mite populations involve several aspects of mite and natural enemy ecology and, possibly, changes in mite or plant physiology.

Delayed resistance to Bt crops: is Arizona's story unique or universal?

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Transgenic cotton producing *Bacillus thuringiensis* (Bt) toxin Cry1Ac that kills pink bollworm, *Pectinophora gossypiella*, has accounted for more than half of Arizona's cotton since 1997. Despite the wide use of Bt cotton and surprisingly high frequency of pink bollworm resistance to Cry1Ac detected in 1997, Bt cotton has remained extremely effective against pink bollworm. Laboratory bioassays show that the frequency of resistance to Cry1Ac did not increase from 1997 to 2003. Field-based estimates also show sustained efficacy during this period. In laboratory-selected strains with up to 3,100-fold resistance to Cry1Ac and survival on Bt cotton plants, resistance is linked with three recessive mutations in the gene encoding a cadherin protein that binds Cry1Ac. DNA-based monitoring of field-sampled individuals using the polymerase chain reaction (PCR) indicates that the frequency of the three identified resistance alleles remains low. Other examples of delayed resistance to Bt crops have been reported from Australia, China, and elsewhere in the USA. Factors delaying pest resistance to Bt crops include refuges of non-Bt crops, low initial resistance allele frequency, recessively inherited resistance, fitness costs, and incomplete resistance. The success of Bt crops exceeds expectations of many, but does not preclude future resistance problems.

Construction of transgenic *Bombyx white egg-1* mutant introduced kynurenine 3-monooxygenase gene

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Bombyx white egg 1 mutant (*w-1*) is characterized by the white colour egg and eye. It was known that the mutant kynurenine 3-monooxygenase gene possesses a partial in the exons. To understand an effect of complementation of wild type *KMO* gene, we introduced *KMO* cDNA with the promoter of native *KMO* or cytoplasmic actin gene (*A3*). We obtained three transgenic lines with A3 promoter and one line with the native promoter. The adult eyes of these lines turned to be brown and the eggs laid by the transformed females were also brown. The analysis by RTPCR showed that the lines with *A3KMO* gene accumulated the transcript in the mid gut, fat body and Malpighian tubes. A line with *KMO* promoter expressed it only in the fat boy and Malpighian tubes. The strength of eye and egg colour in the transgenic line was proportional to the *KMO* expression level. Interestingly, the transgenic larvae with *A3KMO* gene changed the larval skin colour more brownish. These results suggest that the morphological change in *w-1* mutant is caused by a deletion of *KMO* gene. The mutant and *KMO* gene can be used as a host and marker for a construction of transgenic silkworm.

Presence of avidin and aprotinin in the bodies and frass of *Spodoptera litura* (Lepidoptera: Noctuidae) larvae feeding on transgenic tobacco plants.

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To determine whether herbivorous insects and their frass could be environmental sources of novel proteins from transgenic plants, *Spodoptera litura* larvae were fed on transgenic tobacco leaves expressing either the biotin-binding protein, avidin, or the protease inhibitor, aprotinin. Control larvae received non-transgenic tobacco. Samples of 20 larvae each were taken on Days 5, 6 and 7 and two samples of frass on Days 6 and 7. Insects in all treatments grew significantly between Days 5 and 7, but the avidin-fed larvae were significantly smaller and produced less frass than the others on Day 7. All insects produced more frass on Day 7 than on Day 6, but the mass of frass produced per mg insect did not vary from day to day. Both novel proteins were present in larvae at lower concentrations than in the transgenic leaves they had fed upon. Unbound avidin was found in the samples of avidin-fed larvae and their frass. Aprotinin was detected in all larval and frass samples from insects fed aprotinin-tobacco. Unbound aprotinin accounted for only a portion of that found in larvae and in frass.

Bollgard® II cotton, tested for the first time in West Africa, shows excellent lepidopteran control and potential for improved cotton production

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Field studies were conducted in Burkina Faso during the 2003 cropping season to evaluate the efficacy, against lepidopteran insects, of cotton expressing two endotoxins of *Bacillus thuringiensis* subsp. *kurstaki* (Berliner), commonly known as Bollgard® II cotton. Bollgard® II, treated against sucking and piercing pests only, was compared, in two research stations, to local varieties treated against lepidopteran insects and sucking pests. Our data showed that Bollgard® II cotton provided control of major lepidopteran species that attack cotton in Burkina Faso. The average number of bollworm larvae per plant over time was significantly different between transgenic cotton (treated against sucking pests only) and a local variety (treated against lepidopteran insects and sucking pests). The average number of *Helicoverpa armigera* larvae per Bollgard® II plant was reduced by 92% when compared to the local varieties. Bollgard II and the local spraying regimen were similarly highly effective in reducing foliar feeding lepidopterans. The difference in damage by lepidopteran insects, resulted in fewer abscissions of floral and fruiting bodies and higher numbers of bolls per plot in the Bollgard® II cotton. In Frako-Bâ and Kouaré evaluation sites, Bollgard® II cotton rendered a yield 13% and 19% higher, respectively, than the commercial local variety.

Bt crops in Argentina: efficacy and resistance management

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Bt crops began to be de-regulated in 1998 (Bt Cotton: MON 531; Bt Corn: E 176, and MON 810). In 2001, Bt 11 was also authorized. Both Bt crops have steadily increased their acreages. Last season, 45 % of the corn area was planted with Bt corn, and 20% of the cotton was Bt. Bt corn produces Cry1Ab protein, and Bt cotton produces Cry1Ac protein. Corn and cotton producing areas do not significantly overlap, and the only lepidopteran pest in common for both crops in the country is *Spodoptera frugiperda*. Target pests for Bt-cotton are: *Alabama argillacea*, *Heliothis virescens/Helicoverpa gelotopoeon* complex, and *Pectinophora gossypiella*. For Bt-corn, the target species is *Diatraea saccharalis*. The Insect Resistant Management (IRM) group proposes strategies for the management of Bt crops, based on local and external experiences. Research is conducted by third party entomologists from Universities, and National Research Institutes. Mainly, the the strategy is based on a required refuge (non-Bt crop), which is 20% for Bt-cotton, with the possibility of insecticidal-non-Bt applications as needed, according to economic thresholds. For Bt-corn, the requirement is 10% refuge without any insecticides for the target pest. IRM strategy also considers studies such as baselines, emergency plans, monitoring, and continuous studies related to the size of the refuge, that will be discussed during the presentation.

Altered gene expression levels linked to fenvalerate resistance in cotton bollworm, *Helicoverpa armigera*

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The ANO2 strain of *Helicoverpa armigera* is 50-fold resistant to the pyrethroid insecticide fenvalerate; largely due to a single gene, *RFen1*, on Linkage Group 13. The cDNA-AFLP technique was used to search for changes in gene expression levels controlled by or linked to *RFen1*. Larvae from a segregating backcross family were genotyped at a linked marker locus, enabling prediction of their fenvalerate resistance status without exposure to insecticide. mRNA was isolated from individual larvae and used to make cDNA, which was processed for AFLPs (double restriction digestion, ligation to adapters, amplification by two rounds of PCR, and resolution of labelled fragments on a sequencing gel). 256 different primer combinations were tried and a total of 426 bands showing differences between resistant and susceptible groups were identified. Three were cytochrome p450s, with similarity to *Depressaria pastinacella* CYP6AE1 and *Helicoverpa armigera* CYP4S1 and CYP4AP1. The latter was upregulated whereas the other two were down-regulated in the resistant group. In addition, two carboxylesterases were down-regulated and one was up-regulated in the resistant group. Currently, real time RT-PCR is being used to verify the results and full length sequences of these potential detoxification genes are being sought using rapid amplification of cDNA ends (RACE).

Bt: Organic production – new life for the oldest commercial biopesticide.

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The Organization for Economic Cooperation and Development predicts that biopesticide may grow to 20% of the world's pesticide market by 2020. Biopesticides represent approximately 1% of the world pesticide market, and Bt products represent > 75% of biopesticides sold. Other organizations like the World Health Organization, Food and Agriculture Organization and European Commission are also predicting dramatic growth for biopesticides over the next 10-20 years. Sprayable Bt formulations have penetrated cotton, fruit and vegetable, aquatic and other insecticide markets in the US, EU and the world. Organic producer dependence on Bt for worm and, in a more limited fashion, beetle control globally. In fact, many organic workers claim that Bt is more important to the organic community than to other producers because conventional producers have a greater array of management tools to choose from. With organic produce sales increasing steadily worldwide, Bt use and resistance management should be an important consideration in any system that anticipates frequent application, but especially in organic production systems where use can be extreme. Fortunately, the organic formulation of Spinosad™--a fermentation macrocyclic lactone product, is changing management practices in many cropping systems and this may change Bt's organic market dominance. In addition, new insect growth regulators and other novel modes of action may be entering the organic market in the next few years providing new opportunities to implement truly integrated management programs in organic production.

The international arthropod resistance database and newsletter: a report

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A number of resources related to resistance management have been cultivated at Michigan State University in the USA. This presentation will highlight these developments and how resistance workers all over the world can access and participate. The first of these is the Resistant Pest Management Newsletter (RPM News), which has a worldwide circulation of roughly 2,500 resistance workers in academia, industry, and government. It provides information on ongoing changes and advances in the field of resistance management, and is produced in cooperation with the Insecticide Resistance Action Committee (IRAC), the US Department of Agriculture (CSREES) and the US Western Regional Coordinating Committee (WRCC-60). Our goal in implementing a resistance database is to help address arthropod resistance by evolving an integrated, cooperative system to effectively deliver resistance information efficiently. Our information currently resides in an electronic database at Michigan State University's Center for Integrated Plant Systems and is available via the Internet at http://www.cips.msu.edu/resistance/rmdb/. The database design features a central table of 'resistance' records with five key pieces of information: the arthropod, the pesticide, the location (country), the year, and a reference to the document reporting the case. There are secondary tables that elaborate on these five key informational groups as well as auxiliary tables to help refine the information further or link to external data sources.

Juvenile hormone bioanalogs – selectivity, importance and perspectives in insect pest management

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Insect juvenile hormone bioanalogs (JHAs, juvenoids) have been the target of the long-term research in our team for many years. A special interest has been paid to several series of 2-(4-hydoxybenzyl)cycloalkanols for their high biological activity values obtained in screening tests to a broad variety of insect species. Even if a number of these JHAs have been proven to be stable but non-persistent under the environmental conditions, designing of ways for limited increase of their stability in environment and for modification of their physico-chemical properties had been performed. This effort resulted in designing and developing the pioneer hormonogen substances (juvenogens). The juvenogen molecule consists of a JHA, bearing convenient functionality, which can be easily transformed into another functionality by chemical means or by enzymic ways. This step results in a synthesis of a biologically inactive or low active substance, which is characterized by physico-chemical properties different from those found with the parent JHA. The new substance, a juvenogen (a hormonogen chemical species), can be prepared as a targeted species, a substance bearing physico-chemical properties designed in advance. This strategy enables to synthesize "juvenogens upon demand", which are designed for application on selected insect species under selected laboratory or environmental conditions.

UK-wide studies of biodiversity impact of GMHT crops

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By the end of 1998 four genetically modified herbicide-tolerant (GMHT) crops were nearing the end of the regulatory hurdles for commercial growing in the UK. Although human health and direct environmental effects had been assessed and they had been declared safe the question was raised about indirect environmental effects caused by more efficient weed control. The result was the Farm Scale Evaluations, one of the largest agro-environmental impact studies ever undertaken, covering plant and invertebrates at 60-70 field sites for each of 4 crops over a three year period. The background and results from these trials will be discussed in relation to wider issues about insect biodiversity in agricultural landscapes.

Current status and future prospect of Bt cotton commercialization in China

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Transgenic cotton that expressed a gene derived from the bacterium *Bacillus thuringiensis* (Bt) was introduced for combating *H. armigera* in 1997, and rapidly increased to 58% of the total cotton area of 4.8 million ha. in 2003. In the Yellow River cotton-farming region of Northern China, where *H. armigera* has historically caused major yield losses, Bt cotton has been exclusively planted. The pest management tactics associated with Bt cotton planting causes astic reduction in use of insecticides due to the high efficacy of the transgenic cultivars in suppression of *H. armigera* infestations, increases the abundance of beneficial insects and improves the natural control of some pests. The monitoring results indicate that movement toward Bt cotton resistance in *H. armigera* is not apparent in face of intensive planting of Bt cotton over several years, which could be associated with the migratory behavior of the insect and the natural refuges derived from the mixed planting system of cotton, corn, soybean and peanut on single- family holdings with a small scale under one half hectare.

The involvement of microsomal oxidases in pyrethroid resistance in *Helicoverpa armigera* from Asia

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Five contemporary strains of the bollworm *Helicoverpa armigera* Hübner from China, Pakistan and India, all with high resistance to pyrethroids, were compared with a standard susceptible strain that originated from the Cote D'Ivoire in the 1970s ('SCD'). The strong synergism of pyrethroids by PBO implied that an oxidative metabolism could be involved in pyrethroid resistance in all resistant strains. The activities of cytochrome P450 monooxygenases from midguts of final instar larvae to *p*-nitroanisole (PNOD), ethoxycoumarin (ECOD), methoxyresorufin (MROD) significantly increased in all the resistant strains when compared with the susceptible strain. This further implies that cytochrome P450 monooxygenases are involved in pyrethroid resistance in Asian *H. armigera*. Comparative *in vitro* studies of the metabolism of ^{14}C-deltamethrin by midgut microsomes of the resistant PAK and susceptible SCD strains showed that the resistant strain had a much greater capacity than the susceptible strain for the metabolic degradation of deltamethrin. This enhanced metabolic degradation occurred in the presence of NADPH which suggested an oxidative detoxification. In the resistant strains, minor increases in glutathione S-transferase activity (to the substrates CDNB and DCNB), and esterase activity (to the substrate α-naphthyl acetate) further suggested that, of the putative metabolic mechanisms, oxidases are the most important. This study provides the first evidence that cytochrome P450 monooxygenases are a major metabolic mechanism responsible for pyrethroid resistance in *H. armigera* from Asia.

Bt rice studies of ecological risk and nontarget effects in China

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Bt rice has been permitted for field trials since 1998. After then, impacts of our different Bt rice lines containing *cry*1Ab or *cry*1Ab/*cry*1Ac gene with high resistance against rice stem borers and leaffoles, on the arthropod community and population dynamics of some dominant nontarget phytophages and spiders under field conditions was evaluated using different sampling methods. The results showed that our Bt rice lines generally did not have any significant negative effects on the arthropod community in rice paddy habitat, except that sometime resulting in the marked decrease in the abundance of some parasitoids of target pests such as braconids and ichneumonids. These tested lines also did not act any adverse impacts on the predation, growth and development, reproduction of two common spiders, *Pirata subpiraticus* and *Ummeliata insecticeps,* using either leaffolders or planthoppers feeding on Bt rice plants as their prey. In contrast, some Bt rice lines might not be suitable for the brown planthopper, *Nilaparvata lugens* to feed and lay eggs. In addition, no significant adverse effects were found on the survival, growth and development as well as the fecundity of the domestic silkworm, *Bombyx mori* fed with the mulberry leaves contaminated by Bt rice pollen grains.

In vivo effect of PBO on resistance-associated esterases from Australian *Helicoverpa armigera* Hübner (Lepidoptera: Noctuidae)

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Piperonyl butoxide (PBO) is a commercially important and successful insecticide synergist against a variety of insects in house-hold situations and against public health pests. In agriculture use has been restricted to use against the cotton bollworm, *Helicoverpa armigera* (Hübner) (Lepidoptera:Noctuidae). It has been deployed in the field to help partially restore the efficacy of pyrethroids that are currently compromised by metabolic based resistance. It has been established that PBO inhibits some insect esterases associated with insecticide resistance (Gunning *et al.*, 1998; Moores *et al.*, 1998; Young *et al.*, 2002; Young *et al* 2003). Kinetic esterase assays using *H. armigera* reveal that esterase activity was inhibited by the insecticide synergist, PBO, over a 24hr period. Esterase inhibition by PBO did not occur immediately after doseage but occurs with maximum enzyme inhibition being obtained several hours later. Laboratory analysis with *H. armigera* and pyrethroids confirmed that maximum synergism occurred at the pre-treatment times corresponding to maximum *in vivo* esterase inhibition in biochemical studies. These results were confirmed in the field when using a 5 hr pre-treatment of PBO prior to pyrethroid exposure. These results indicate that with improved temporal application of PBO, it may be possible to restore some pyrethroid efficacy in *H. armigera*. It would also be expected that partial restoration of efficacy with other conventional insecticides currently compromised by esterase based resistance mechanisms would occur.

PTD significantly synergize Bt and overcome the low resistance of diamondback moth to Cry1Ac

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Protein Transduction Domains (PTD) are kinds of peptides which can automatically penetrate bio-membrane and be acted as a carrier to transfer foreign molecule into cell(Science. 1999,285(3):1569-1572.). A fusion gene,pET21b-TAT-Cry1Ac, consisting of Cry1Ac and TAT(a kind of PTD) ,as well as its control pET21b-Cry1Ac was constructed and expressed in E.coli. The toxicity of this two expressed and purified fusion proteins to Diamondback moth were determined. To the susceptible strain the LC50 of Cry1Ac and TAT-Cry1Ac are 20.55 mg/L and 4.25 mg/L; and 124.23mg/L and 12.81 mg/L to the low resistant strain. TAT-linked Cry1Ac show a synergistic ratio of 4.84 folds to the susceptible strain, 9.70 folds to the resistant strain, and restore the susceptibility of low resistant Diamondback moth to Cry1Ac. PTD show strong potential in synergizing Bt insecticidal protein and overcoming Bt resistance.

Fluctuating asymmetry of target and non-target insects in response to the stress of GM cotton

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Fitness and fluctuating asymmetry (FA) in response to the stress of GM cotton were examined among the different symmetric characters of target and non-target pests (cotton bollworm, *Helicoverpa armigera*, CB, and cotton aphid, *Aphis gossypii*) and a beneficial species (Asian lady beetle, *Harmonic axyridis*, ALB). The results indicate that non-target effects do arise from some GM cultivars. The FA values of CB in lab decreased gradually generation by generation. The light-trap catches of CB collected in northern China with different planting history of Bt-cotton (1~5 years) also exhibited a similar trend on FA. Bt-cotton and conventional cotton are equally suitable hosts for cotton aphid. The fitness and FA of ALB and cotton aphid were similarly when fed on the aphids from both Bt-cotton and conventional cotton. But the Bt+CpTI cotton present significant impacts on the fitness of cotton aphid that their intrinsic rate of increase and fecundity decrease significantly in the first and second generation. The aphids feeding on Bt+CpTI cotton showed higher FA and their feeding behavior detected by EPG is different significantly with Bt-cotton and conventional cotton. The possibility of FA as a sensitive indicator for the ecological risk and environmental stress of GM crops are discussed.

"P450 from multiple insect genomes: from phylogeny to function"

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"Cytochromes P450 are enzymes involved in the metabolism of signal molecules such as ecdysteroids, juvenile hormones and pheromones, and in the response to chemical stresses of the environment (insecticides, plant secondary compounds). P450 are encoded by genes from a very large family, the CYP genes. *Drosophila melanogaster* has 85 CYP genes with 5 pseudogenes (http://P450.antibes.inra.fr). In the mosquito, *Anopheles gambiae*, only 10 P450 gene orthologs were found vs the predicted ±40. Results on the P450 genes from the genomes of *Drosophila pseudoobscura*, *Apis mellifera* and *Bombyx mori* will be presented. The current assembly of the honeybee genome reveals significantly fewer P450 genes in this insect when compared to the three Diptera and the silkworm. Mitochondrial P450 genes of insects are particular when compared to mitochondrial P450s of vertebrates, because some of them (CYP12) behave like environmental response genes (multiple genes, no 1:1 orthology between species, inducibility) whereas others are involved in steroid hormone metabolism (the Halloween genes). Microsomal P450 genes involved in conserved, endogenous functions can also be identified by orthology, following their characterization in one species. The case of the P450 involved in juvenile hormone biosynthesis is an example that will be discussed.
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Insecticide resistance and its management in the cotton bollworm *Helicoverpa armigera* in Pakistan

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The cotton bollworm, *Helicoverpa armigera* (Hubner) (Lepidoptera: Noctuidae) is a key pest of cotton and other crops in Pakistan. Indiscriminate use of pesticides prompted the development of insecticide resistance in this pest. Resistance to pyrethroids has been on the rise since 1991. Resistance factors are now >100 to all the pyrethroids. Endosulfan resistance remained moderate to high. However, some thiophosphates and carbamates mostly exhibited low levels of resistance. We never ran out of options for the control of *H. armigera*. The latest introduction of new chemistries with novel modes of action added to the effective armory. An IRM strategy including other IPM tactics has been recommended to the cotton growers for the better control of *H. armigera* and other cotton pests.

Mutations in acetylcholinesterase (AChE) of the cotton melon aphid (*Aphis gossypii* Glover) and the peach potato aphid (*Myzus persicae* Sulzer) confer resistance.

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Two mutations in the *ace1* gene of *Aphis gossypii* are associated with insensitivity of acetylcholinesterase (AChE) to carbamate and organophosphate insecticides. The first of these, S431F (331 in *Torpedo californica* numbering), is associated with insensitivity to the insecticide, pirimicarb. The mutation to a phenlyalanine is consistent with a loss of sensitivity to pirimicarb in a range of both *A. gossypii* and *M. persicae* clones. A rapid RFLP diagnostic allows the identification of individuals of both aphid species with a resistant genotype. This diagnostic further reveals the presence of S431 in several other pirimicarb-susceptible aphid species. The serine at this position in the wild-type enzyme has only been reported for aphids and is considered the reason why pirimicarb has a specific aphicidal action. A second mutation, A302S (201 in *T. californica*), confers insensitivity to a wide range of carbamates and organophosphates and has, to date, only been found in resistant strains of *A. gossypii.*

Stem application of monocrotophos in okra: dissipation of monocrotophos residues

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The status of residues of monocrotophos at harvest and its dissipation pattern in okra fruits when employed as stem application at the rate of 1:6 (one part of monocrotophos in six parts of water) were studied in a field experiment. The residues of monocrotophos were below detectable level (BDL) during first picking when the stem application was imposed at or before 38 days after germination (DAG). The repeated application, even during fruiting, evidently established the cumulative residues of monocrotophos in okra fruits. The residues (0.57 ppm) recorded in okra fruits on first day of stem application accumulated up to 3rd day (2.21 ppm) and dissipated gradually to maximum residue level (MRL) of 0.2 ppm with Ttol. of 11.27 days. Therefore stem application of monocrotophos in okra should be restricted to 12 days prior to picking of fruits.

Effects of cypermethrin and chlorpyrifos residues in cattle dung on the development of the dung beetle *Copris tripartitus* (Coleoptera: Scarabaeidae)

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Fresh dung was collected from untreated cattle and cattle dosed with a spray-on formulation of cypermethrin and chlorpyrifos (2.1g /cow) on days 1, 3, 5 and 7 post-treatment. The dung was bioassayed using the dung beetle *Copris tripartitus* Waterhouse. Residues of cypermethrin and chlorpyrifos were sufficient to inhibit oviposition by *C. tripartitus* in day 1 dung but there was no significant effect on egg laying in dung collected at days 3-7 post-treatment in sexually matured female. But, in the second generation residues of cypermethrin and chlorpyrifos were sufficient to inhibit oviposition by *C. tripartitus* in treatment after 1 and 3 days. Although there was no significantly different effect on egg laying in dung collected at days 3-7 post-treatment in first breeding but, in second breeding, toxic effect reveal in 3 and 5 days dung treatments. Nulliparous dung beetles fed for 200days on dung collected 0, 1 and 3 days after treatment of cow with cypermethrin and chlorpyrifos show little ovarian development.

Effect of ecdysone agonists on tortricid male/female orientation

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The attractiveness and responsiveness of adult codling moth, *Cydia pomonella* (L.), redbanded leafroller, *Argyrotaenia velutinana* (Walker), and obliquebanded leafroller, *Choristoneura rosaceana* (Harris), exposed to surfaces treated with the ecdysteroid agonist methoxyfenozide were investigated in wind tunnel and orientation tube assays. When males were exposed to water-treated surfaces for 48 h the mean percent recaptures of such treated males in the wind tunnel assay were almost always (depending on species) significantly greater than the recaptures of males that had been exposed to methoxyfenozide-treated surfaces. Similarly, in the orientation tube assay, methoxyfenozide-treated males almost always had significantly lower mean levels of individuals exhibiting sexual excitability (intense wing fanning and upwind orientation) and mean distances traveled upwind in the orientation tubes. The two assays demonstrated that males exposed to methoxyfenozide-treated surfaces were not as responsive to calling females (treated and non-treated) as were the non-treated males. Conversely, females exposed to methoxyfenozide-treated surfaces were just as attractive to non-treated males as were water-treated females. It appears that a male's ability to respond to a calling female is more negatively affected by the ecdysone agonist than a female's ability to call and attract males.

Assessing the role of cytochrome P450s in insecticide resistance of *Lucilia cuprina*, *Helicoverpa armigera*, and *Drosophila melanogaster*

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Cytochrome P450 are heme containing enzymes that perform a diverse array of REDOX reactions against exogenous and endogenous compounds. Characterised insect genomes have approximately 90-100 individual cytochrome P450 genes and the biological function for most of these is unknown. Previous studies have implicated the over-expression of some cytochrome P450 genes in insecticide resistance via metabolic degradation. Using degenerate PCR and cDNA library sequencing, we have cloned and characterised a number cytochrome P450 genes from the Australian sheep blowfly (*Lucilia cuprina*) and the cotton bollworm (*Helicoverpa armigera*), two insect pests of Australia. We have generated a micro-array of these cytochrome P450 genes, and are characterising the expression of these genes at different life cycle stages and in different tissues. We are also using - micro-array and realtime quantitative PCR to compare the expression levels of cytochrome P450 genes between resistant and susceptible strains. Our studies in *L. cuprina* and *H. armigera* are complemented by studies in *osophila melanogaster*. Using a micro-array containing all 90 *D. melanogaster* cytochrome P450 genes, we are studying gene induction by insecticides. We are over-expressing induced cytochrome P450 genes, and determining if over-expression can confer insecticide resistance.

Novel genetic basis of field-evolved insect resistance to Bt toxins

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Insecticidal toxins from *Bacillus thuringiensis* (Bt) are widely used in sprays and transgenic plants to control insects that attack crops and transmit diseases, but evolution of pest resistance threatens their continued efficacy. Diamondback moth (*Plutella xylostella*) is the only pest that has evolved Bt resistance in open field populations, although several others have been selected for resistance in the laboratory or glasshouses. The most common type of resistance (Mode 1) is characterized by recessive inheritance, >500-fold resistance to at least one Cry1A toxin, and negligible cross-resistance to Cry1C. The primary mechanism of Mode 1 resistance is reduced binding of Bt toxins to midgut membrane target sites. Mutations affecting a Cry1A-binding midgut cadherin protein are tightly linked to laboratory-selected Mode 1 resistance in two major pests, *Heliothis virescens* and *Pectinophora gossypiella*. The cadherin gene has thus been considered the prime target for DNA-based screening for resistance. Here we show that field-evolved Mode 1 resistance in diamondback moth has a different genetic basis, indicating that screening for resistance in the field should not be restricted to searching for cadherin mutations.

Managing pest resistance to Bt crops in India

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Bt transgenic cotton first introduced in India in 2001 was cultivated on over 100,000 hactares in 2003. Besides, other Bt transgenic crops such as rice, egg plant, cole crops, pulses are in various stages of development. The target pest for Bt cotton is the American bollworm, *Helicoverpa armigera* and for Bt rice it is yellow stem borer (YSB), *Scirpophaga incertulas*. Based on the baseline toxicity data against the former in India and estimation of frequency of resistance alleles in the populations of YSB in the Philippines, it appears that these two species can rapidly buildup resistance against the Bt endotoxins. Bt resistance management for transgenic cotton is made mandatory requirement for the growers who have to maintain 20% refuge crop or minimum of 5 rows around the Bt cotton field, whichever is larger. However, in view of inadequate knowledge and expertise among the farmers, small land holdings and apprehension of expected reduction in yield due to non Bt lines, farmers tend to ignore the refuge cultivation. Studies on the baseline susceptibility of the American bollworm to Cry1Ac have helped in the monitoring of resistance in the Bt cotton. Similar studies are being done in other crops too. We feel, Bt resistance management should include detailed studies on various characteristics of Bt resistance in local pest populations of the target pests, and explore possibilities of field level management in tandem with the local agronomic practices and environmental conditions.

Bt maize effects on butterflies.

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Transgenic plants have been widely adopted by U.S. farmers despite concerns about potential nontarget impacts of this technology. The possibility that maize (*Zea mays*) genetically engineered to express *Bacillus thuringiensis* endotoxoin for control of European corn borers (*Ostrinia nubilalis*) may have impacts on non-target lepidopterans through pollen drift onto neighboring non-agricultural plants has received considerable attention. Since initial concerns about Bt corn were raised by a laboratory study, additional studies both in the field and the laboratory have revealed the remarkable complexities of accurately assessing risks of pest management practices in agroecosystems. These studies are examined in the context of ecological, economic, social, and cultural factors that affect food production and pest management in the United States.

The relative fitness of Bt resistant and susceptible *Helicoverpa armigera* **– implications for the refuge strategy**

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Transgenic crops producing the Cry1Ac toxin of *Bacillus thuringensis* (Bt) was introduced in Australia, China and India primarily for the control of *Helicoverpa* spp. As *Helicoverpa armigera* has a demonstrated capacity for developing resistance to many synthetic insecticides, development of resistance to Bt toxins remains the biggest threat to the sustainability of transgenic technology. The refuge strategy has been widely adopted to counter the onset of resistance. This strategy is most effective when resistance is conferred by rare, recessive alleles and when mating of heterozygotes in prevented. Under these conditions, the evolution of resistance can be substantially delayed. However, we have found evidence that two of these conditions are not always met. Results from glasshouse experiments have demonstrated that, although resistance in *H. armigera* is functionally recessive on young Cry1Ac cotton, attenuation in toxin expression results in survival of heterozygous genotypes on late season Cry1Ac cotton. Our resistant strain of *H. armigera* takes longer to develop on cotton that expresses high levels of Cry1Ac than their susceptible counterparts on non-Bt cotton. However, loss of potency in older cotton plants mitigates asynchrony in late season insect populations. The implications of these findings for resistance management of Bt crops will be discussed.

Novel approaches for controlling mosquitoes using natural inhibitors for trypsin modulating oostatic factor

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Trypsin Modulating Oostatic Factor (TMOF) is a mosquito decapeptide that modulates trypsin biosynthesis in mosquitoes, flies and *Heliothis virescens*. Feeding the hormone and its analogues to mosquito and *H. virescens* larvae stopped trypsin biosynthesis in their midgut by binding a specific gut receptor and controlling the translation of the trypsin message. Cytoimmunochemical analyses show that the hormone is not only synthesized in the ovary as was initially hypothesized, but is also found in the central nervous system cells of adults and larvae. Since the hormone affects the growth and development of mosquito larvae the hormone and several analogues have been cloned and expressed in chlorella, yeast (*Saccharomyces cerevisiae* and *Pichia pastoris*) and Alfalfa cells to control insects using biorational insecticides. These results and the effect of the transformed cells on larval growth and development will be discussed.

Fluorometric PCR applications to insecticide resistance analysis in disease vectors.

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As insect genomes are published, molecular techniques have begun to be widely applied for the detection and assessment of insecticide resistance. Vectors develop insecticide resistance through mutations of insecticide target sites, such as the voltage-gated sodium channel, and increased activity of detoxification enzymes such as oxidases and esterases. The ability to use preserved or desiccated field-collected insects makes molecular analysis of insecticide resistance an especially attractive option. A useful approach to measuring all of these mechanisms is fluorometric PCR. This method allows detection of resistance mechanisms based on point mutations at the insecticide target site, resistance gene amplification, or increased expression of resistance genes. Knowledge of resistance gene sequence is a prerequisite in the development of these methods. Our efforts to apply quantitative PCR to detection, measurement and assessment of insecticide resistance in an assortment of disease vectors are summarized. The same molecular methods that serve to detect resistance are also central to the determination of population genetic relationships of the resistance alleles. We discuss our present state of knowledge of molecular markers and insect genomes as they relate to insecticide resistance.

Tri-trophic prey-mediated impacts of insect-resistant tobacco on a predatory carabid beetle

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Larvae and adults of a New Zealand endemic predatory beetle, *Ctenognathus novaezelandiae*, were fed an exclusive diet of prey (*Spodoptera litura*) reared on control tobacco, or transgenic tobacco expressing either a protease inhibitor (PI) or a biotin-binding protein (BBP). BBP prey-fed beetle larvae accumulated greater mass than the other 2 treatments over a 7-week period in the middle of the 14-week experiment. Over the entire experiment, the mean biomass of BBP-fed larvae was greater than that of PI- but not control-fed larvae. There were no differences among treatments in larval survival or quantity of prey consumed. Adult female beetles grew larger and survived longer than males in all 3 treatments. Combined gender adult mean biomass over 40 weeks was the same for all 3 treatments. However, control-fed females had greater total biomass than PI or BBP-fed females, but control-fed males had less biomass than males given the other treatments. Over the entire experiment, BBP-fed adults consumed slightly less than the other 2 treatments. Fecundity was low in all 3 treatments and there were no differences among them. The greatest adult differences detected were between genders rather than among treatments, where analysis of data collected weekly showed only minor and transient effects.

Analysis of acetylcholinesterase insensitivity in California insect pests

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Acetylcholinesterases are important members of the serine hydrolase super-family of enzymes and are the targets of the organophosphate (OP) and carbamate insecticides. Although the future use of these insecticides in California is somewhat uncertain, due to the FQPA, they are still widely used in agriculture, both as stand-alone insecticides, and as synergists of the pyrethroid insecticides. As a consequence of their widespread and varied usage, OP and carbamate resistance has been identified in many important pests. Acetylcholinesterase insensitivity is an important mechanism of resistance to these insecticides, and cross-resistance within these chemical groups is not uncommon. We have conducted extensive studies on resistance in several important California insect pests. In this presentation, we will describe our studies on two pests – the beet armyworm (BAW), *Spodoptera exigua*, and the glassy-winged sharpshooter (GWSS), *Homalodisca coagulata*. The BAW has been long-established in California agriculture, whereas the GWSS is a recent introduction. These two pests differ in their pesticide exposure histories and this seems to be a contributing factor in their relative susceptibilities to OPs and carbamates. The effectiveness of OPs and carbamates for the management of these pests is discussed based on our toxicological and biochemical studies.

Activity profiles of neonicotinoid insecticides in plants and implications for IPM

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The duration that an insecticide residue is active in a crop in terms of lethal and sub-lethal effects on insect populations is a critical concept in pest management that has been poorly developed. Application rates, the timing and spacing of applications, and the choice of materials could all be influenced by more reliable information on the activity period of insecticides in crops. Commercial ELISA kits were used to determine concentrations of imidacloprid and thiamethoxam in citrus, grapes, and melons at regular intervals following soil treatment. The uptake and residual profiles differed significantly for both compounds in all crops. Peak concentrations of imidacloprid in xylem fluid of large citrus trees were not attained until 6-8 weeks after application compared to 2 weeks for thiamethoxam. However, near-peak levels of imidacloprid were sustained for 6-10 weeks before declining gradually in contrast to thiamethoxam titres (applied at ¼ the rate of imidacloprid) that declined rapidly. The same pattern of higher application rates and longer residual of imidacloprid was also observed in melons. Comparison of young vs. old leaves on primary branches of melon vines revealed much higher concentrations in older leaves, a counterintuitive finding because better whitefly control is attained on younger leaves.

Contamination of refuges by Bt toxin genes from transgenic corn

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Transgenic crops producing insecticidal toxins from *Bacillus thuringiensis* (Bt) are widely used to control pests, but their benefits will be lost if pests evolve resistance. The mandated high-dose/refuge strategy for delaying pest resistance requires planting refuges of toxin-free crops near Bt crops to promote survival of susceptible pests. We report that pollen-mediated gene flow up to 31 m from Bt maize caused low to moderate Bt toxin levels in kernels of non-Bt maize refuge plants. Immunoassays of non-Bt maize sampled from the field showed that the mean concentration of Bt toxin Cry1Ab in kernels and the percentage of kernels with Cry1Ab decreased with distance from Bt maize. The highest mean Bt toxin concentration in kernels of non-Bt maize plants was 45% of the mean concentration in kernels from adjacent Bt plants. Most previous work on gene flow from transgenic crops has emphasized potential effects of transgene movement on wild relatives of crops, lanaces, and organic plantings, while implications for pest resistance have been largely ignored. Variable Bt toxin production in seeds of refuge plants undermines the high-dose/refuge strategy and could accelerate pest resistance to Bt crops. Thus, guidelines should be revised to reduce gene flow between Bt crops and refuges.

Insect comparative genomics: a carboxyl/cholinesterase perspective

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Our comparative genome analyses of principle enzyme families involved in metabolic resistance to insecticides have generated new research opportunities. The data provide a credible basis for identifying the resistance-associated enzymes of insects and the means for identifying previously unknown molecules involved in fundamental biological process such as development. Carboxyl/cholinesterase enzymes have contributed to a major part of insect biochemical and genetic research. The most notable member of this supergene family, acetylcholinesterase (AChE), is the key processing enzyme for the neurotransmitter acetylcholine. Many other esterases have now been implicated in the metabolism of specific hormones and pheromones and accordingly in various aspects of insect development and behaviour. Intriguingly, the carboxyl/cholinesterase family provides a striking example how gene duplication/radiation/deletion shape the evolution of novel biology, notably how the function of a catalytically active domain has evolved to mediate receptor-ligand interactions that regulate morphogenetic process during development of the nervous system. We have recently classified carboxyl/cholinesterases into three principle functional groups (intracellular enzymes associated to dietary detoxification; secreted enzymes including those that process pheromones and hormones; receptors and enzymes involved in neuro/developmental functions). Our data (gene inventories) are also providing a template for new studies concerning the honeybee and silkworm genomes.

Characterization of CTCF proteins in mosquitoes

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CTCF (CTCCC-binding factor) is a multivalent, multifunctional transcription factor well characterized in vertebrates. Of particular interest is its association with every known vertebrate insulator and its emerging role in epigenetic regulation. Endogenous mosquito insulators would be valuable elements to flank transgene insertions, inhibiting both position effect and position effect variegation, thus allowing for more consistent gene expression. CTCF-like cDNAs were cloned, expressed and characterized from the medically important mosquitoes *Aedes aegypti* and *Anopheles gambiae.* The gene is transcribed constitutively in all developmental stages and appears to be up-regulated both in the ovaries from blood-fed females and in early embryos. Putative binding sites for the *An. gambiae* protein were determined *in-vitro* using PCR-assisted binding site selection and confirmed in gel-shift assays. ChIP (chromatin immunoprecipitation) assays are currently in progress. Additionally, an RNAi experiment to knockdown expression both in *An. gambiae* adult females and in *osophila melanogaster* females and early embryos, followed by RNA isolation and hybridization to microarray chips is being performed to identify specific CTCF targets. The results of these experiments will be presented.

Esterase mediated cross resistance between pyriproxyfen and buprofezin in B-biotype *Bemisia tabaci* in Australia

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B-biotype *Bemisia tabaci* was first detected in Australia in 1994. B-biotype *B. tabaci* is a severe pest worldwide and poses a serious threat to many ornamental, agricultural and horticultural industries in Australia. Control of B-biotype *B. tabaci* is hampered by resistance to many conventional insecticides. Consequently, insect growth regulators such as buprofezin and pyriproxyfen are important insecticides in managing this pest. Field resistance has been detected to both insecticides with cross resistance between the two as well. Studies have shown resistance to buprofezin in B-biotype *B. tabaci* to be esterase mediated and experiments were designed to investigate the potential of pyriproxyfen to bind to *B. tabaci* esterases. Esterase inhibition experiments showed inhibition by pyriproxyfen was greater in the buprofezin resistant strain than in its unselected counterpart. Concentrations as low as 0.321nM pyriproxyfen gave 70-80% inhibition of resistant *B. tabaci* esterases. Cross resistance between IGRs with different modes of action poses a grave threat to the viability of the management strategy of silverleaf whitefly in Australia.

Efficacy of novaluron for Colorado potato beetle control and potential for resistance development.

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Novaluron (RimonÒ 10EC), a novel insect growth regulator, could play a key role in development of practical integrated pest management (IPM) programs for Colorado potato beetle (CPB), *Leptinotarsa decemlineata* (Say). In laboratory tests, novaluron exhibited potent insecticidal activity against CPB larvae (e.g. 2nd instar (L2) LC_{50} = 0.42 ppm). Furthermore, the insecticidal activity of a single novaluron application was highly persistent under field conditions. In bioassay, 65-85% of L2 from an insecticide-susceptible laboratory strain died when fed potato foliage treated with novaluron at 25 and 50 g a.i./ha, respectively, 28 days after application. Thus, the selection pressure for novaluron-resistant CPB may be intense long after population densities have been reduced below an economic threshold level. Studies were also conducted to identify geographic variability and potentially problematic CPB populations before the widespread use of novaluron in the field. The susceptibility of L2 to novaluron was determined for 27 different populations collected from 6 Canadian provinces in summer of 2003. Despite no previous exposure to novaluron, the populations differed considerably in susceptibility with mortalities at a diagnostic dose ranging from 55-100%. Our results suggest that despite the potential of novaluron in CPB management, the possibility of rapid development of resistance exists.

Bemisia tabaci, **biotypes and insecticide resistance management.**

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Many insect species exist as a complex of biotypes that can be differentiated by a variety of genotypic or phenotypic parameters. Some of these are of ound importance in pest management. For example some biotypes of the cotton whitefly (*Bemisia tabaci*) appear to be associated with different ecological, behavioural and insecticide-resistant characteristics. The processes that mediate the balance and evolution of such co-existing forms are neither well-described nor understood. It is clear however, that in some instances, the evolution of biotypes among pest species is influenced by the tools used for their control (e.g insecticides). In an effort to understand the factors that influence the dynamics of co-existing biotypes and how these impact upon the implementation of management strategies and the dynamics of insecticide resistance, we examine Mediterranean populations of *Bemisia tabaci*. We present work that characterises insecticide resistance in recently collected strains of 'B' and 'Q' biotypes, and examine the ability of these forms to introgress (by observing courtship behaviour, mating success, hybrid sex ratio and hybrid fecundity). We examine the consequences of such introgression for the inheritance of insecticide resistance.

Hydolysis of organophosphorus and pyrethroid insecticides by insect carboxylesterases modified by *in vitro* mutagenesis

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Resistance of *Lucilia cuprina* to Ops is due to mutations in the gene encoding carboxylesterase E3 that enhance its ability to hydrolyse insecticides. Mutagenesis *in vitro* and expression of these modifications (G137D and W251L) have confirmed their role. These and nearby amino acids were substituted by others expected to affect the efficiency of the enzymes in hydolysing OPs and pyrethroids. Various substitutions of W251 had a broadly similar effect to W251L on OP hydolysis, but some enhanced malathion carboxylesterase. With the exception of F309L, nearby mutations generally diminished OP hydolysis. W251L was the most effective substitution to improve permethrin hydolysis (up to 130-fold) whilst decreasing the trans:cis hydolysis ratio to favour the more insecticidal isomer Other mutations around the catalytic site generally enhanced pyrethroid hydolysis relative to the wild type, the most effective being F309L, which enhanced transpermethrin hydolysis more than W251L. Complementary assays with individual enantiomers of the dibromo analogue of cispermethrin showed the wild type and most mutants to have a marked preference for the least insecticidal 1S conformation of the cyclopropanecarboxylate, but this was reversed in mutants with the F309L substitution. The results show the potential to design such recombinant enzymes for bioremediation of insecticide residues.

Enhancing biodiversity in sugar beet through appropriate management of GMHT crops

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The UK Farm Scale Evaluations of the environmental impact of genetically modified herbicide-tolerant (GMHT) crops reported that the biodiversity of wildlife in GMHT beet was adversely affected compared to that in the conventional crops. This was attributed to better control of weeds by glyphosate, to which the GM beet was tolerant, with knock-on consequences for the invertebrates that depended on those weeds for survival. In other separate studies on sugar beet, alternative glyphosate programmes, involving band sprays and/or delayed sprays (compared to the aft label recommendations) resulted in much weedier plots than under conventional regimes at different times in the season, with consequent benefits for some invertebrates. In many cases this was achieved with no loss of yield compared to conventional systems. Another regime that relied on one early spray of glyphosate resulted in greater numbers of late germinating weeds than in conventional sytems, and consequently greater seed production by those weeds for the benefit of seed-eating beetles and autumn-feeding birds. GMHT technology with regard to beet is so flexible that almost any scenario can be created and still allow farmers to grow a itable crop.

Ecdysteroid agonist insecticides – mechanism, selectivity and importance in IPM

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Non-steroidal ecdysone agonists belonging to the bisacylhydrazine class of compounds are a new generation of insecticidal compounds that cause premature lethal molts in susceptible intoxicated insects. Of these, three bisacylhydrazine compounds, tebufenozide (MIMIC®, CONFIRM®, ROMDAN®), methoxyfenozide (INTREPID®, RUNNER®, PRODIGY®, FALCON®) and halofenozide (MACH 2®) have been commercialized. Both tebufenozide and methoxyfenozide are predominantly toxic to lepidopteran pests of cotton, corn and other agronomic plants. Halofenozide, a soil insecticide, is toxic to both lepidopteran and coleopteran pests. All three insecticidal compounds manifest their toxic action in susceptible insects via interaction with the ecdysone receptor, which is also the target of the insect molting hormone, 20-hydoxyecdysone. This selective toxicity of both tebufenozide and methoxyfenozide to lepidopteran pests is largely due to their high binding affinity to EcRs from those insects. The toxic action of halofenozide, which is a weak binder of EcR, is primarily due to its relatively higher metabolic stability in susceptible insects. The mode of action of the three insecticides at the whole insect, ultra-structural and biochemical levels will be discussed and data shown to demonstrate their safety to beneficial insects, mammals and the environment. The use of these insecticides in IPM and IRM programs will be emphasized.

Prodigy™ - a new IPM-compatible product for insect control in Australian crops

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Methoxyfenozide (formulated as Prodigy™ 240 SC and previously coded as RH 2485), is a moult-accelerating compound (or MAC), from the same chemical family as tebufenozide (Mimic™ 700 WP), a product which has been in the Australian market for many years. Methoxyfenozide is generally active at lower rates and has a wider spectrum of activity than tebufenozide, even though activity of both is strictly limited to the Order Lepidoptera. This selectivity makes methoxyfenozide an ideal candidate for use in integrated pest management (IPM) programmes as it has no direct effect on non-lepidopteran predators and parasitoids. As the only MAC compound used in Australia against helicoverpa, with no documented field resistance methoxyfenozide is a valuable new tool for inclusion in helicoverpa resistance management strategies. It's introduction reduces selection pressure on older products and further delays the onset of resistance. Development to date has focused on helicoverpa control in cotton and tomatoes, but methoxyfenozide does have several exciting possibilities. Globally, methoxyfenozide is already recognized as having excellent activity against *Spodoptera* spp. and may prove to be a cost-effective alternative for control of armyworms in a range of Australian crops (e.g. pastures, sugarcane, cereals and turf).
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Gene resources for insecticide discovery

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The majority of insecticides and nematicides in current use act on only a small number of target receptors. Drawing on the resource provided by complete genome sequences, we have sought to identify and validate novel targets for nematicides and insecticides. The excitatory glutamate receptors involved in signalling at the insect neuromuscular junction are an attractive, but poorly understood candidate target. We have identified new members of this gene family from whitefly *Bemisia tabaci* and the bollworm *Helicoverpa armigera*. The availability of these gene sequences provided new insight into the diversity of excitatory glutamate receptor sub-types in insects. Pharmacological and molecular genetic approaches were used to identify novel serotonergic (5-HT$_3$) receptors regulating pharyngeal pumping in the free living nematode *Caenorhabditis elegans*. Lead compounds were assessed for their toxicity to *C. elegans*, *Myzus persicae*, *H. armigera* and *Heligosigmoides polygyrus*. A number of *C. elegans* cDNA sequences, one or more of which are believed to encode subunit(s) of the nematode's pharyngeal 5-HT$_3$ receptor were isolated. Preliminary gene knockdown data indicates that one of the cloned receptor subunits is vital for the proper development and normal pumping of the pharynx. Our data provide the first evidence for the existence of excitatory, 5-HT$_3$-like, receptors in nematodes and the first indication that receptors of this type are valid targets for pesticides.

Molecular mechanisms of phosphine toxicity and the genetics of resistance

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Phosphine (PH_3) is the only globally significant fumigant available to protect stored grain from insect infestation. Phosphine is a somewhat hydophobic reducing compound that can complex with biologically significant metal ions, partially disrupt mitochondrial function and induce cytotoxic levels of oxidative stress. Despite a complex array of potential modes of action, strong resistance (600X) to this compound in the pest insect, *Rhyzopertha dominica*, is achieved with resistance alleles at only two loci, neither of which negatively affects fitness. Phosphine resistant mutants generated in the model organism, *C. elegans*, have an unusually long lifespan. One of these mutants is completely resistant to the toxic effect of hyperoxia in conjunction with an otherwise sublethal exposure to phosphine. Mutant and wild type nematodes exhibit no differences in terms of phosphine induced changes to either mitochondrial morphology or ATP levels. The mutant, however, appears to be protected from phosphine induced oxidative damage to lipids. Surprisingly, genome-wide microarray analysis in *C. elegans* does not implicate mitochondrial dysfunction or oxidative stress as the primary causes of phosphine toxicity. Rather, the stress response appears to mimic the expression profiles associated with the lifespan extension mutant, *daf-2*, as well as the unfolded protein response induced by denatured proteins.

Pyrethroid insecticide resistance and treated bednets efficacy in malaria control

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Metabolic and k resistance mechanisms were characterized in An. stephensi and this easily colonized species was used to evaluate the efficacy of ITNs. Bioassays performed on 4th instar larvae showed a resistance ratio of 182. The resistance was partially suppressed by PBO (54%) and DEF (51%) or both (68%). Biochemical assays revealed that esterases, monooxigenases and glutathione S-transferase levels were 1.72, 1.86 and 1.88-fold respectively in the DUB-S compared to the susceptible strain Beech. The overall permethrin metabolism rates by DUB-S mosquitoes measured by HPLC were 1.5-fold more than by Beech strain. The classic k mutation was detected in II6 of the para-type sodium channel gene from DUB-S strain. Bioassays performed on the resistant DUB-S mosquitoes using different pyrethroid-treated bednets showed that except for deltamethrin and a-cypermethrin treated bednets, the rest of the tested pyrethroid-treated bednets at standard WHO recommended doses did not produce adequate mortality rates. The deterrency test performed on susceptible and resistant An. stephensi showed that there are significant differences between the entry rates of susceptible and resistant mosquitoes into the exposure tube containing permethrin treated bednet. These data show that when mosquitoes have both k-type and metabolic resistance mechanisms, the efficacy of pyrethroid-treated bednets is questionable.

Update on the honey bee genome project

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Along with their longstanding importance for agriculture, honey bees (*Apis mellifera*) have been important models for behavior, physiology, ecology, and genetics. They play a considerable role in evolutionary comparisons across insects and in attempts to understand the forces that lead to, maintain, and challenge insect societies. A aft sequence for the honey bee genome was funded in 2002 by the U.S. National Institutes of Health, and Department of Agriculture, following a 'white paper' competition to identify compelling eukaryotic sequencing targets. The project began that year at the Baylor College of Medicine Human Genome Sequencing Center (www.hgsc.bcm.tmc.edu), with input from individual scientists and the Sanger/Wellcome Trust. The current assembly (version 1.1, January 2004) reflects 1.25 Gbp of mainly short-insert (shotgun) sequences, giving an estimated 6.0-fold coverage of the 210 Mbp genome. The assembly is comprised of 11,600 scaffolds (N_{50} = 223 kbp) containing 30,074 contigs (N_{50} = 19 kbp). Approximately 95% of known EST's, cDNA's and markers are in this assembly. Current plans are to incorporate reads from an AT-biased library and an existing BAC library and to generate cDNA's useful for gene calling. An updated assembly and an initial gene list are anticipated by the time of this meeting.

EST analysis of genes expressed in the different tissues of the spruce budworm, *Choristoneura fumiferana*

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More than 35,000 sequence reads have been generated from five cDNA libraries of the epidermis, midgut, fat body, whole larvae and cell line of the spruce budworm, *Choristoneura fumiferana*. These expressed sequence tags (ESTs) have been assembled into approximately 8000 unigenes using Phrap program. Annotation of these sequences has been conducted against the protein database Swiss-Prot/TrEMBL and the GenBank of NCBI. The unique sequences have been classified for their molecular function, cellular location, and biological process using the Gene Ontology classification system. A spruce budworm EST database has been established and is available to the scientific community. These annotated EST sequences will serve as a resource base for gene mining, comparative analysis and genome mapping. The ESTs have also being used for developing cDNA microarrays, which will be used to study the global gene expression in the spruce budworm in response to different stresses. This project is supported by funds of Genome Canada and Canadian Biotechnology Strategy.

Implementation and impact of transgenic Bt cottons in Australia

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Bt cottons (INGARD®) expressing the CryIAc endotoxin, were commercialised in Australia in 1996/97 through an industry agreed deployment strategy which limits use to 30% of total cotton. A comprehensive management strategy minimises the risk of Bt resistance in *Helicoverpa armigera*, the target pest. In 6 years of commercial use Bt cottons have reduced pesticide applications for *Helicoverpa* by an average of 50%, providing major environmental benefits. Fibre quality has not been affected, while in some cases yields have increased. Despite early difficulties with variable performance, growers have rapidly adopted the technology and will gain long-term benefits through enhanced environmental sustainability, as Bt cotton is used in environmentally sensitive areas. Perhaps more importantly Bt cottons show great potential for IPM systems where a combination of soft approaches yields considerable benefits. That single gene Bt cotton has not been a "magic bullet" is fortunate in demonstrating that Bt cottons need to be introduced within an IPM system and supported by rigorous resistance management strategies. Two-gene Bt cottons expressing Cry 1Ac and Cry 2Ab (Bollgard II) have now been commercialised and will replace INGARD® in 2004/5. BGII provides enhanced efficacy and further reductions in pesticide requirement with associated environmental benefits. BGII also bring reduced resistance risk, but will remain part of a conservative resistance management strategy similar to that for INGARD varieties.

Pleiotropic effects of insecticide resistance genes in an aphid pest on vulnerability to attack by parasitoid wasps

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Studying the phenomenon of insecticide resistance provides a rare and valuable insight into the fundamental processes underlying evolution as resistance often appears over short timescales in human terms. In effect, this process represents a classic case of 'forced evolution' and as a result can produce resistance mechanisms that have negative pleiotropic effects on fitness in the absence of insecticides. The paper describes experiments done at different spatial scales measuring the vulnerability of insecticide-susceptible and –resistant peach-potato aphids (*Myzus persicae*) to attack by the parasitoid, *Diaeretiella rapae*. It also discusses the potential impact of the findings on Integrated Pest Management strategies utilising beneficial insects.

Insect AChE

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Two genes encode acetylcholinestesterase (AChE) in insects, but only one seems to be expressed in the synapse of the central nervous system. Protein is a dimer anchored to the presynaptic memebrane via a glycolipid. Structure has been solved for the *Drosophila* enzyme, the active site is buried 20 Å inside the protein, at the bottom of a narrow gorge. This structure results to complex kinetics resulting from the traffic of substrate and products in the active site gorge. Another consequence of this structure is the sensitivity to insecticide: insect AChE is more sensitive to insecticide than its vertebrate counterpart. This property has been used to design biosensors to detect insecticide residues and the enzyme has been engineered to improve its capacity of detection. Extensive use of insecticides resulted in insecticide resistance. Several mutations have been evidenced. They often combined in the same site resulting in an enzyme more resistant and with a broader spectrum of resistance. Mutations affected enzyme efficiency : they decrease or increase the limitating step of substrate hydolysis, deacetylation and decreases enzyme stability. This alteration of enzyme has been compensated in most of strains by an overproduction of the enzyme.

First season of methoprene application mirroring Fire Ant Control Centre treatment program has not had any prompt deleterious effects on the native soil invertebrate assemblage.

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In response to the accidental introduction of the invasive ant *Solenopsis invicta* (Buren) to Brisbane, Australia, an eradication program was mounted. This program employs a range of chemicals including the insect growth regulators methoprene and pyriproxifen, which though specific to invertebrates and intended solely for *S. invicta*, may have deleterious effects on other invertebrates. The impact of Methoprene on soil invertebrates and nutrient cycling is being evaluated using a control-impact study conducted in eucalyptus woodlands where *S. invicta* is absent. Methoprene is applied to treatment sites following Fire Ant Control Centre procedure, with 3 applications of 2 kg/ha each treatment season (November to April). Soil samples are collected: before treatment; and one month after each subsequent treatment over two treatment seasons (Nov 2002-May 2004). Soil invertebrates are extracted and sorted to morphospecies to form a community dataset for each site. Due to their numerical dominance, and also importance for processes such as decomposition and nutrient cycling, only the mite component of the invertebrate community will be reported here. Results at the completion of the first treatment season indicate that the application of Methoprene is not associated with a decrease in any of the diversity parameters. Community data are also analysed using ordination techniques to investigate whether the actual species composition is influenced by methoprene application.

Large-insert BAC libraries for fundamental studies in Lepidoptera

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The tobacco hornworm, *Manduca sexta*, the tobacco budworm. *Heliothis virescens*, and the Müllerian mimic, *Heliconius erato*, are well-established lepidopteran models for studying fundamental problems in olfaction, neurobiology, and development (*M. sexta*), evolution of insecticide resistance and plant host specialization (*H. virescens*), and wing pattern mimicry (*H. erato*). We have constructed two separate BAC libraries for each species using *Bam* HI and *Eco* RI partial digestion. Average insert sizes of *Bam* HI and *Eco* RI libraries are 152 kb and 165 kb for *M. sexta*, 171 kb and 150 kb for *H. virescens*, and 175 kb and 153 kb for *H. erato*, with <5% non-recombinant clones. Genome coverage totals an estimated 12.9x for *M. sexta*, 16.3x for *H. virescens* and 15.4x for *H. erato*. We are screening the libraries with well-conserved single-copy genes to better estimate genome coverage and obtain BAC clones to use as anchor loci for comparative studies. All libraries are publicly available at http://hbz.tamu.edu. We are indebted to L. Riddiford, F. Gould, and O. McMillan for insects, and M. Cornwall and N. Pierce for vouchering specimens. Supported in part by NSF grant no. IBN-0208388 to MRG, HBZ, and CW.

Functional genomics of the midgut of the cotton bollworm, *Helicoverpa armigera*

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The Lepidopteran insect, *Helicoverpa armigera,* or cotton bollworm, is a major pest of agriculture worldwide. Many control strategies involve orally-transmitted control agents that target the larval midgut. A functional genomics study of this organ has the potential to increase understanding of its molecular and cellular biology and to identify new targets for pest control. We have undertaken an EST project to identify genes important for midgut function, growth and development and gene/enzyme pathways involved in insect-plant and insect-pathogen interactions. Sequence analysis of midgut cDNA libraries now covers ~5,000 ESTs each from young (2nd and early 3rd instar) and from 5th instar larvae. Clustering yielded a unigene set of ~3,900 contigs. Searches against the UniProt (EBI) database and model organism proteomes yielded confident Gene Ontology annotation for ~55% of the contigs. These include genes associated with key midgut functions: digestive hydolases; nutrient and ion transport and pH maintenance; a range of detoxification enzymes and genes forming the peritrophic membrane or involved in midgut cell proliferation, differentiation and adhesion. Many have not previously been identified in lepidopteran systems. Additionally, comparison with silkworm ESTs identified likely orphan lepidopteran genes. Microarrays based on the non-redundant gene set are being established for expression iling studies.

Pesticide resistance in Brazil and IRAC-BR activities: current situation and perspectives

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Recognition of pesticide resistance problems in Brazil dates of the late 1960s, but it was not followed by further studies until the 1990s, when the interest was renewed due to reports of insect-pest control failures. The establishment of the Brazilian Insecticide Resistance Action Committee (IRAC-BR) in 1997 was a reflex of this concern and provided new impetus to pesticide resistance studies in the country. The major activities of the IRAC-BR have been centered on institutional, research and educational programs. The institutional program is based on the promotion of the Committee and its activities, while the research program establishes priorities and provides support for projects of interest. At present, the research effort is centered in two long-standing projects, *Tuta absoluta* in tomatoes and *Spodoptera frugiperda* in maize/cotton, and several other shorter-term projects. The educational program provides basic training in Insecticide Resistance to a diverse public. Annually, IRAC-BR offers short-courses, workshops, lectures and round-table discussions in different regions of the country. IRAC-BR also sponsors a National Research Award, in two categories, undergraduate and graduate research projects. This award was created to stimulate creativity, positive criticism and human resource formation in Insecticide Resistance Management in Brazil. These initiatives from IRAC-BR have been well received in Brazil and the fruits from this effort are beginning to ripen.

Esterases mediate resistance to biological insecticides in *Helicoverpa armigera*

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The cotton bolloworm *Helicoverpa armigera* is a serious pest of cotton and other summer crops in Australia, where it has a long history of insecticide resistance to most insecticides. Particularly critical for Australian agriculture was the evolution of pyrethroid resistance. Pyrethroid resistance which was found to be largely due to overproduction of specific esterase isoenzymes (R_m 0.24 - 0.33), which sequester and hydolyse pyrethroids. Modern pest resistance management strategies have incorporated new biological insecticides with novel modes of action. Such biological insecticides are spinosad and *Bacillus thurlingiensis* toxins and as well insecticidal proteins of B thurlingiensis which have been incorporated into genetically engineered plants. Nonetheless, there is an enduring threat of resistance. *H. armigera* has already developed resistance to spinosad and some CRY toxins and in this paper we report esterase mediated resistance mechanisms.

A microsomal esterase confers insecticide resistance in *Spodoptera littoralis*

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Spodoptera littoralis (Lepidoptera: Noctuidae) (Boisduval), the Egyptian cotton leafworm, is a major pest of cotton and vegetable crops in Mediterranean Europe and Africa. A field strain collected from cotton at Adana in Turkey was found to have a resistance factor of 8-fold against chlorpyrifos by leaf dip bioassay. Preliminary studies of AChE sensitivity against chlorpyrifos-oxon indicated no evidence of insensitive AChE. An enhanced microsomal esterase was revealed that correlated with the observed resistance when compared to a susceptible standard strain. We report here on the purification and kinetic parameters of this microsomal esterase.

Transgenic corn and rootworm: ecology, evolution and management.

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The western corn rootworm in North America depends almost exclusively on corn for larval survival and adult nutrition. Two-year crop rotations were very effective at controlling rootworms since oviposition historically only occurred in corn. In recent years however, crop rotation has failed in the corn-belt east of the Mississippi river. Adults in this region lay eggs in fields other than corn and because of rotation these eggs hatch in corn the following year. The change in the behavior of adult rootworms resulted in the increased use of insecticides and a renewed interest in developing ecologically sound control strategies. We report on a series of modeling efforts that examined how land use lead to the evolution of behavioral changes in adults and how landscape diversity and weather patterns contribute to the dispersal and ultimate distribution of this rotation resistance variant in North America. We then examine a variety of control options combining cultural practices with transgenic crops and other crop technologies to simultaneously manage rotation resistance and the risk of developing resistance to transgenic corn. The results were sensitive to landscape diversity, toxin dose, expression of resistance, initial allele frequency and density-dependent larval survival.

EST-cDNA clones were mapped based on RFLP in the silkworm, *Bombyx mori* L.

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We have developed the genetic analysis by using molecular markers based on restriction fragment length polymorphism (RFLP) in the silkworm, *Bombyx mori*.. In the silkworm there is no crossover on female chromosomes, so a (RF02 x RF50) female was crossed to a RF02 male and from this BC_1 segregant more than 250 ug individual DNAs were prepared and used to know their genotypes. Every clone gave homozygous or heterozygous genotype for each individual and the patterns for each clone of 15 BC_1 segregants could be identified from each other. After the 28 patterns for every chromosome were made, every new clone was easily to know their linkage group from the genotyping of 15 individuals. After linkage analysis, all clones were mapped by using the 100 segregants of the cross between a (RF02 x RF50) male and a RF02 female. From all of the genotypes, non-recombinant individuals are those with either all A, homozygous, or B, heterozygous, but recombinant individuals have both A and B. The linear order of clones is known after data is sorted and distance between clones determined by counting the recombinants. Now about 300 cDNA clones have been mapped on molecular genetic map.

Molecular tools for reconstructing the past and predicting the future of insecticide resistance

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We have been using the tools of modern biotechnology to reconstruct the past and predict the future course of the evolution of metabolic resistance to organophosphorus (OP) insecticides in the higher dipterans *Lucilia cuprina* (Calliphoridae) and *Musca domestica* (Muscidae). Resistance in both species is due to mutations that convert a carboxylesterase enzyme into a catalytically inefficient but physiologically protective phosphotriesterase. There are a pair of allelic mutations in each species which give resistance to structurally distinct sets of OPs. The two mutations involve the same substitutions in the same residues of orthologous proteins in the two species. In order to reconstruct the history of the resistance we have been using PCR to clone and sequence the gene involved from preserved museum specimens over an 80 year temporal transect. This transect goes back over 20 years before the first use of OP insecticides. We find that at least one of the resistance alleles existed in natural populations before the introduction of the pesticides. In order to predict the future course of resistance we have been using the tools of directed *in vitro* evolution and rational enzyme design in attempts to improve the catalytic efficiency of the extant natural mutants. We have now been able to recover *in vitro* mutants which are catalytically superior in the degradation of OPs.

The management of cotton aphid (*Aphis gossypii* Glover) in Australian cotton

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Until recently, cotton aphid, *Aphis gossypii* Glover, has been a persistent but readily controlled secondary pest of Australian cotton. However, during the 1998-1999 cotton-season, control failures linked to resistance were reported for organophosphate and carbamate insecticides, with some populations controllable only with diafenthiuron. Since then resistance hotspots have developed annually over eastern Australia that have only been controllable with new chemistries. The reasons for this are complex and include seasonal differences in rainfall patterns affecting over wintering host availability for aphids and a reduction in insecticide use, particularly endosulfan, with the coincident introduction of transgenic cotton. In response, aphid management has been multi-faceted and based on a complex integrated pest management (IPM) approach. This includes an Integrated Resistance Management Strategy (IRMS) that has recently been altered to better manage the selective aphicide pirimicarb. This useful IPM chemical is usually quickly lost to high-level target site resistance but we aim to manage it through improved tactical product use.

Chemical control of western flower thrips in Australia

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Insecticides are the main method used by commercial growers to control Western flower thrips (WFT) *Frankliniella occidentalis*, **in Australia. However, as has occurred overseas, WFT are resistant to a number of insecticides and insecticide groups. Furthermore, the current field application rates of many insecticides used in Australia may be too low to be effective. This would increase selection for resistance, because insecticides are being applied at sub-lethal doses. To test this, we carried out a series of field trials. Crops tested included ornamentals, tomatoes, lettuce, strawberries, cucumber and capsicum. Six key chemicals (abamectin, acephate, dichlorvos, endosulfan methidathion and pyrazophos) were evaluated. All field trials were integrated with insecticide resistance monitoring to remove the confounding effects of chemical resistance. Field trials confirmed that many application rates used for WFT were too low to be effective. Resistance monitoring showed that abamectin, methiocarb, and pyrazophos are the only chemicals where resistance was not detected. Low-level resistance was detected against acephate, dimethoate, endosulfan, fipronil, methamidophos, methidathion and spinosad. Increasing levels of resistance were detected against chlorpyrifos, dichlorvos and maldison.**

The GMO guidelines project - developing the science in risk assessment of transgenic plants

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The GMO Guidelines Project (www.gmo-guidelines.info) has developed a conceptual framework for a methodology of case-specific ecological risk assessments of genetically modified plants. It consists of a generic framework that is constructed on five scientific sections and incorporates many different scientific disciplines: 1) Problem Formulation and Options Assessment (PFOA), 2. Transgene Expression and Locus Structure (TELS), 3. Biodiversity and Non-target Impacts, 4. Gene Flow and its Consequences, 5. Resistance Evolution and Management. The generic framework of the GMO-Guidelines is comprised of a risk assessment model, which is driven by systematically structured, detailed scientific questions. The model can be applied to any case and the information and data for answering each question tailors the assessment to the specific context yielding a case- and country-specific risk assessment. We applied the framework to three case examples: Bt-maize/Kenya, Bt-cotton/Brazil and Bt-cotton/Vietnam. In our presentation, we will briefly present the major outcomes of these case studies. Several unique features of the Project: a) We work collaboratively with scientists from developing countries and jointly conduct the case study risk assessments. Thereby, we build scientific capacity and build confidence in their own scientific capabilities; b) We frame risk assessment in a multi-stakeholder deliberative process of PFOA.

Modification of sucrose synthesis in GM aspens: effects on leaf-beetle preference.

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GM forest trees potentially pose ecological problems beyond those faced by transgenic agricultural crops because: forest trees are essentially undomesticated which makes escape of transgenics to the wild more probable and trees also have long life spans, dominate the forest ecosystems and define the structure of many communities. Therefore, it is of outmost importance that we gain basic knowledge of the potential ecological effects of transgenic tree species. The aim was to determine if genetic modifications of aspens, not primarily intended to influence natural enemies, will lead to changes in the plants nutritional quality and secondary chemistry, thus effecting interactions between natural enemies (insects) and GM trees. We used GM (*Populus tremula x tremuloides*) aspens modified with respect to increased sucrose-synthesis (ZEA SPS), resulting in increased mesophyll sucrose concentration, dry mass and growth. Two different lines (different degrees of up regulation) were used and unmodified wild-type aspen served as controls. We used two common leaf feeding chrysomelid beetles, *Lochmeae caprea* and *Phratora vitellinae* in cafeteria test. *Phratora vitellinae* utilized transgenic aspens marginally less than the wildtype whereas no difference was found for *Lochmeae caprea* preference. The relationship between these results and plant primary and secondary chemistry and its implication for plant-herbivore interactions is discussed.

Biorational insecticides – important components in pest management of cotton, vegetable and ornamentals, an overview

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There have been efforts to develop biorational insecticides with selective properties to act on biochemical sites present only in the target arthropods and not in non-target organisms. This approach has led to the development of insecticides such as the neonicotinoids (which affect nicotine acetylcholine receptors), the benzoylphenyl ureas (BPUs) and buprofezin (which inhibit chitin synthesis), and the juvenile hormone (JH) mimics and ecdysone agonists. Compounds originating from natural products, which act on GABA gated chloride channel, glutamate receptors and the chloride channels (such as the avermectins and the spinosyns) and at the mitochondrial level (chlorfenapyr) with much more association to the insects than mammals, have been developed and used successfully to combat diverse insect pests and spider mites. Additional new groups of insecticides include the phenylpyrazoles (fipronil), the pyridines (pymetrozine) and the oxadiazines (indoxacarb), which act on GABA receptors, on the nerves controlling the salivary pump and on the sodium channel, respectively. All the above, are considered biorational, or "low risk" insecticides, which have relatively low detrimental effect on the environment and its inhabitants, rendering them important potential components in IPM programs. The selectivity of these biorational insecticides to beneficials and resistance evolution in key pests will be discussed.

Selection of insecticide resistant strains of African malaria vector mosquitoes

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Insecticide resistance in the major African malaria vector mosquitoes seriously impacts on control operations in many countries. In 1999/2000, South Africa experienced its worst malaria epidemic in over 50 years, due to the return of pyrethroid resistant *Anopheles funestus*. The situation is confounded by cross-resistance to carbamates in *An. funestus* and low level resistance to DDT and pyrethroids in the other southern African vector *Anopheles arabiensis*. Resistant strains of both species have been established and laboratory investigations into the mechanisms of the resistance carried out. Only a metabolic mechanism appears to be responsible for the pyrethroid resistance in *An. funestus*. In *An. arabiensis*, however, two mechanisms have been found in populations from different parts of Africa, one a knockdown (*kdr*) type resistance and the other a metabolic mechanism. The possibility of cross-resistance to other classes of insecticides is discussed.

Actions of clothianidin and related neonicotinoids on recombinant and native nicotinic acetylcholine receptors

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Imidacloprid and related neonicotinoids are insecticides acting selectively on insect nicotinic acetylcholine receptors (insect nAChRs). Among neonicotinoids, imidacloprid has pyridine and imidazolidine moieties, whereas clothianidin lacks both moieties. Since it was conceivable that such structural differences could result in different actions of these two insecticides on nAChRs, we have investigated, using two-electrode voltage-clamp electrophysiology, the agonist actions of clothianidin and imidacloprid on *Drosophila* SAD-chicken b2 hybrid nAChR expressed in *Xenopus laevis* oocytes. Also, we have used whole-cell patch-clamp electrophysiology, to study clothianidin actions on native nAChRs in the TAG neurons from male adult American cockroaches as well as in the mushroom body neurons from *Drosophila melanogaster* larvae. Clothianidin was found to activate not only the recombinant SADb2 hybrid nAChR but also native insect nAChRs with a greater efficacy than that of imidacloprid. Structure-agonist action relationship analysis suggested that opening of the imidazolidine ring is likely to account for the greater agonist efficacy of clothianidin compared to that of imidacloprid.

Novaluron (Rimon), a novel IGR – cross-resistance, selectivity and importance in pest management

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Novaluron, 1-[chloro-4(1,1,2-trifluoro-methoxyethoxy)phenyl]-3-(2,6-difluorobenzoyl)urea, a novel benzoylphenyl urea acts by both ingestion and contact. It is a powerful suppressor of lepidopteran larvae such as *Spodoptera littoralis* and *Helicoverpa armigera* (by ingestion) and whiteflies such as *Bemisia tabaci* (by contact). The compound has translaminar activity thereby affecting leafminers such as *Liriomyza huidobrensis*. Its residual activity under field conditions ranges between 10 and 30 days depending on environmental conditions. Artificial rain at a rate of 40 mm/h applied 5 and 24 h after novaluron treatment in a cotton field had no appreciable effect on the potency of novaluron on *S. littoralis* larvae. Novaluron has no appreciable cross-resistance with the juvenile hormone mimic pyriproxyfen or with the neonicotinoids. Hence these compounds can be used as components in insecticide resistance management for controlling whiteflies and other insect pests in cotton and field crops. Novaluron has no appreciable effect on parasitoids and phytoseiid mites, and probably has a mild effect on other natural enemies. Hence, it can be considered an important component in IPM programs.

The susceptibility of *Aedes aegypti* from northern Queensland to synthetic pyrethroids

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Aedes aegypti is the only mosquito vector of the dengue virus in northern Queensland. Despite complete reliance upon synthetic pyrethroids to control adult *Ae. aegypti* during dengue outbreaks, little is known regarding the susceptibility of *Ae. aegypti* from northern Queensland to these insecticides. Insecticide exposure bottle assays and biochemical microplate assays were conducted to examine the susceptibility of *Ae. aegypti* to synthetic pyrethroids. The susceptibility of five strains of *Ae. aegypti* from northern Queensland to five commonly used synthetic pyrethroids was compared with that of an insecticide-susceptible laboratory strain. All strains tested were fully susceptible to the synthetic pyrethroids used by mosquito control authorities in northern Queensland (lambda-cyhalothrin and deltamethrin). However, some wild strains showed significantly reduced susceptibility to permethrin, bioresmethrin and alpha-cypermethrin. Biochemical microplate assays were used to measure the activity levels of two types of enzymes (β-esterases and monoxygenases) that often confer resistance to synthetic pyrethroids. Elevated esterase activity was detected in three wild strains relative to the reference strain, but elevated oxidase activity was not detected in any of the mosquito strains. The presence of resistant traits in wild populations of *Ae. aegypti* demonstrated here supports the need to establish a resistance monitoring programme in northern Queensland to ensure the continued success of dengue outbreak control.

Identification and sequence analysis of the chromosomal region containing *nsd-2*, a densovirus-resistance gene in *Bombyx mori*

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Bombyx mori densovirus (BmDNV) multiplies in the columnar cell nuclei of the mid gut epithelia in the silkworm, *Bombyx mori*. It is classified into 2 types, DNV-1 and DNV-2. Some silkworm strains were identified as resistant against DNV-1 and/or DNV-2. So far four non-susceptibility genes have been reported, namely; *nsd-1*, *Nid-1*, *nsd-2* and *nsd-Z*. However, none of them have yet been isolated as responsible genes. Studies on these genes are useful in understanding the mechanism of non-susceptibility in relation to the viral invasion and multiplication. We identified four closely linked cDNA markers with *Nid-1* and *nsd-2*. By taking advantage of *Bombyx* genome information, the BAC contigs covering these cDNA markers were expanded by screening of the BAC library with BAC end sequences, fingerprinting and shotgun sequencing of BAC clones. At the same time, backcrossed F_1 segregants were selected by virus nonsusceptibility and the linked boundary of the chromosome was defined by sequence analysis.

The use of surface plasmon resonance in characterisation of metabolic resistance

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Insecticide resistance can be the result of target-site insensitivity, metabolic resistance, or both. In general, target-site insensitivity can be readily determined by direct assays on the receptor or target involved. Metabolic resistance can often be harder to characterise, often being implicated indirectly by synergism studies or the use of model substrates. These may or may not accurately reflect the 'true' interaction between pesticide and the degradative enzyme. The use of BIAcore surface plasmon resonance technology allowed the direct interaction of insecticides with a purified esterase to be measured. Additionally such interactions are measured in real time, allowing a unique insight into the binding between resistance-associated esterase and xenobiotics.

Bt-maize and *Trichogramma* wasp – two compatible strategies in control of European corn borer (*Ostrinia nubilalis*)

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The efficacy of *Bt* maize and Trichogramma wasp (Tw) against *O. nubilalis* was tested in two field trials in 2002 and 2003. The hybrids of maize MON 810 (Bt-maize), Monumental (isolinie) and Raissa (local variety) were used. Tw was applied as preparation Trichocarp. Plant injury before harvest on untreated plot ranged from 0.75 to 2.5 per plant in 2002, and 0.22 – 1.4 in 2003. Biological efficacy of *Bt* maize was 100%, the biological efficacy of Tw ranged from 35 to 69%. The increase of yield on *Bt* maize plot reached 21 - 27% in 2002, and 7 – 15% in 2003 and on plots treated by Tw ranged from 0.6 to 17 in both years. The content of four mycotoxins in grain of maize ascertained by ELISA will be presented. Mathematical model of population dynamics of *O. nubilalis* in strips of maize treated by Tw in comparison with untreated strips will be presented. Tw can be applied in insect resistant management (IRM) of European corn borer. Strips of non-transgenic maize hybrid protected by Tw can be used as a refugee to decrease spread of Bt-resistance. The work was funded by the Ministry of Agriculture of the CR project no. QD 1360.

Fluorescence PCR techniques for detecting mutations in the sodium channel gene and for analysis of family 6 oxidase genes conferring pyrethroid resistance in *Aedes aegypti*

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Multiple mutation sites in transmembrane segment 6 of domain II of the sodium channel gene confer pyrethroid resistance in *Aedes aegypti*. Existing strategies for identification of genotypes using PCR are often difficult to use with routine success, especially in the case of heterozygotes. We sought to develop more robust PCR methods based on fluorometric PCR and primer mismatch PCR that remove much ambiguity in scoring results. Here we describe several approaches to making the scoring of resistance genotypes a more straightforward and reproducible procedure. Data are shown for *Aedes aegypti* populations from several continents expressing varying degrees of pyrethroid resistance, as well as for the two mutations known from *Anopheles gambiae*. Elevated expression of P450 oxidases can also confer pyrethroid resistance in *Aedes aegypti* populations. A conserved repeat motif in the downstream P450 reductase binding site of *Aedes aegypti* family 6 P450 genes was used as a probe for quantitative analysis of oxidase copy number and expression. Oligonucleotides were selected for conserved regions of sequence (across the *Culicidae*) and for specific oxidase sequence variants. Through fluorescence PCR analysis, this allowed a comparison of oxidase copy number and expression as well as comparison to homologous sequences in other *Aedes*, *Culex*, and *Anopheles* species.

Biokinetics in insecticide discovery

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Biokinetics is the study of uptake, distribution, metabolism and excretion of xenobiotics and is an important factor to address in the development of new chemical insecticides. Methods have been developed within Syngenta to address these factors on whole insect systems, and the plants upon which insects feed, to gain an insight into biokinetic processes. An understanding of these factors allows iterative cycles of chemical synthesis to improve biokinetic properties which ultimately lead to increased biological potency. Furthermore, biokinetics has played a major role in understanding the translation of in vitro biochemical signals to in vivo biological potency. Conversely this can occur the other way round when a propesticidal compound is tested and again biokinetics can be used to ascertain the conversion of the parent to the active primary metabolite. This was recently exemplified in cyanotropane insecticidal chemistry which demonstrated potent biological activity on sucking pests (hemiptera) but had very weak micromolar activity on the insect nicotinic acetylcholine receptor (nAChR). Biokinetics proved the rapid conversion of the parent to a stable metabolite in insects which had high nanomolar affinity to the insect nAChR thus linking the in vivo to in vitro potency of this chemical series.

Genetic variation and phenotypic plasticity in *Leptinotarsa decemlineata* under different selection pressures

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Invasive pest species provide us an opportunity to study genetic variation linked to selection pressures in natural populations. As pests are invading new areas they face new temperature regimes as well as human induced selection by pesticides. Different countries use also different varieties of cultivated crop species, which may retard insect performance. All these selection pressures are expected to shape the life history traits of a species. Finland is still free of the Colorado potato beetle *Leptinotarsa decemlineata* but on the boarder of its range. To assess beetles potential to invade Finland we compared the life-history traits of four European populations (Italy, Poland, Estonia, Northern Russia). Adult size varied very little between populations when reared on two different temperature regimes. However, developmental times differed among populations suggesting that there may be enough genetic variability for the beetle to adapt further to cooler climates. The frequency of alleles conferring resistance to permethrin was highest in Polish, and declined for Estonian and Russian populations, which suggests that the Polish population has experienced highest pesticide selection. However, resistance to aziphos-methyls was in overall high and similar in all populations. Finally, host plant variety affected little the adult size and the developmental time.

Insecticide choice in alfalfa hay may protect water quality

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Some insecticides used for controlling Egyptian alfalfa weevil, *Hypera brunneipennis,* have been detected in California's surface waters and are of concern due to their impact on water quality and toxicity to some aquatic life. To assess the impact of insecticide choice on water quality, we collected tail-water samples from on-farm alfalfa sites in the northern Sacramento Valley, CA over a 3-year period. Samples were collected during irrigation after organophosphate and pyrethroid sprays were applied. We found significant differences between insecticide classes in the mortality of *Ceriodaphnia dubia* (water flea) a test organism used to detect pesticides in water. Nearly all sites where organophosphate insecticides were used resulted in 100% water flea mortality in a 24-hout test of tail-water samples. The pyrethroid-treated sections of the same fields exhibited insignificant water flea mortality nor were any pyrethroids found to a detection limit of 50 ppt. The pyrethroids used in this study provided significantly better control of Egyptian alfalfa weevil than the organophosphates with no significant differences in beneficial insect counts. Although water runoff does not always occur in alfalfa fields, insecticide choice may be an important tool for protecting water quality in this crop.

Resistance traits of genetically modified silver birch (*Betula pendula*) against insects

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Genetic modification has recently been applied to forest trees, aiming to improve productivity, quality and resistance to insect pests and diseases. We introduced a sugar beet chitinase gene into silver birch, using the *Agrobacterium* mediated gene transfer. Chitinases have been characterised as defense related proteins, particularly in plant/fungus interactions. Fifteen transgenic birch lines showing varying levels of sugar beet chitinase 4 expression and a wild type birch were applied in a field test. Birch seedlings were planted in an agricultural field according to a randomized block desing in 2000. We conducted an inventory of spatial and temporal distribution of insect species and damages caused by herbivorous insects on birch seedlings. Systematic sampling of transgenic and wild type trees was carried out early, middle and late in the growing season 2003 using four randomized field trial blocks. Furthermore, we studied the effect of the quality of partially defoliated birch foliage on performance of *Orgyia antigua* (Lymantridae) larvae in the laboratory. Transgenic and wild type trees were artificially defoliated in nine field trial blocks. This study has been a part of a risk assessment project aiming to provide new information on the biological interactions between genetically modified trees and other organisms.

Effects of neem (*Azadirachta indica* A. Juss) on rice bug, *Leptocorisa chinensis* (Dallas)

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Ovicidal, ovipositional, nymphicidal effects of neem (*Azadirachta indica* A. Juss) on rice bug, *Leptocorisa chinensis* (Dallas), were tested under laboratory and caging conditions. Neem oil alone and azadirachtin (Aza) at 60-180 ppm did not show ovicidal effect on D7. However, a high mortality was observed on D10 in treatment with the mixture of neem oil and abamectin at a low rate and moderate mortalities in treatments of Aza at high rates > 180 ppm. Neem extracts did not significantly affect the survival of adults and the damage to rice when used as ovipositional deterrents. Treatment of 1-3 instars showed that the mixture of neem oil and abamectin achieved the best results in increasing the mortality and delaying the nymphal development, followed by treatments of Aza in a rate-dependent manner. Toxicity of Aza to various instars was also nymphal-age and observation-time dependent. No second instars in rice survived to three weeks when two sprays of Aza at 60 ppm were applied. Two sprays of Aza at 60 ppm and 1.4% neem oil significantly increased rice yield by 23-44%. Egg number was moderately reduced on Aza treated plants. Neem may be integrated into an existing IPM package for *L. chinensis*.

Overview on environmental impact of Bt-corn (Event Bt 176) on non target organisms

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Field studies were conducted in Italy to assess the potential impact of transgenic *Bacillus thuringiensis* corn on non target organisms. Bt corn was modified to express the Cry1Ab toxin against the lepidopteran pest *Ostrinia nubilalis.* We monitored above-ground herbivores, predators, and parasitoids as well as soil-related organisms such as Carabidae and Nematoda. Community structures and degree of biodiversity were compared between transgenic and isogenic crops. No significant impact of Bt-corn on plant-dwelling organisms was observed. However, remarkable differences were found in the case of *Lydella thompsoni*, a parasitoid of *O. nubilalis*. Contrasting results were detected for soil organisms. The Carabidae assemblages were similar, and any differences appeared to have no correlation with Bt maize. The biodiversity of the Nematoda was quite low for both types of crops and no significant differences were found for the majority of the cases. However, Nematoda assemblages were significantly different in Bt than isogenic crops planted in clay-rich soils. These results indicate that we should not neglect taxa of agro-ecosystems before releasing transgenic crops or any other xenobiotics.

Increased efficacy of conventional insecticides against greenhouse pests by the use of piperonyl butoxide (PBO).

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The Tobacco Whitefly, *Bemisia tabaci* (Gennadius) and Western Flower Thrip, *Frankliniella occidentalis* (Pergande) are serious insect pests of agricultural and horticultural crops throughout the world. They can cause damage through both direct feeding and also through transmission of virus. The primary strategy for control for both species remains chemical sprays. However, both have the potential for fast development of resistance due to short generation time, high fecundity, and a haplodiploid breeding system. As a result, high resistance has evolved to conventional insecticides. Results of *in vitro* assays indicated efficacious use of piperonyl butoxide could substantially reduce or nullify the metabolic resistance mechanisms conferring resistance in these species. Subsequent field trials where a pyrethroid (lambda cyhalothrin) and an Insect Growth Regulator (pyriproxyfen) were applied following a pre-treatment of piperonyl butoxide, reflected the *in vitro* findings.

Biofumigation: plant cellular disruption and water achieves high toxin levels in soil

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Some brassicas are potentially large sources of isothiocyanate (ITC) toxins, in the same chemical family as the methyl isothiocyanate produced by the widely used broad-spectrum pseudo-fumigant pesticide metham sodium. Endeavours to harness this potential in the form of biologically active green manures for IPM is dubbed 'biofumigation'. Recent laboratory studies have shown that ITC concentration in soil increased 100-fold with a combination of disruption of plant tissue at the cellular level (achieved by freezing and thawing) and high levels of moisture. Analytical determination of ITCs in soil following manipulation of *Brassica* green manure crops showed that similar improvements in the concentration of these compounds could be achieved in the field. It was crucial to thoroughly pulverise plant tissue with a club-type mulcher running at high speed. A blade mulcher tended to chop the plant tissue into fragments without liquefying it. Leaving the pulverised mulch on the surface and immediately watering it heavily achieved the greatest ITC concentration in soil. It is evident that the intrinsic water content of brassicas is insufficient to allow maximum hydrolysis of the precursor glucosinolates to ITCs. Addition of water boosts this process and carries the ITCs into the soil.

The crucial role of calcium in enhanced biodegradation of soil-applied pesticides

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Enhanced biodegradation occurs where particular, but otherwise entomologically innocuous, soil bacteria are selected by repeated use to catabolise soil-applied pesticides. It is a subtle process akin to resistance in reducing soil-borne pest control efficacy. It is more widespread than is generally recognised, possibly because of its indirect mode of action, counter-intuitive mechanism and probable confusion with resistance. Extremely severe enhanced biodegradation of the widely used broad-spectrum soil-applied pesticide metham sodium occurs in Australia in intensive horticulture, particularly on sandy soils. Highly resilient spore-forming bacteria are the cause. Pesticide persistence in affected soil was just seven hours, but 15 days in similar soil never exposed to metham sodium and in affected soil sterilised by autoclaving. A positive correlation has often been reported between the risk of onset or degree of enhanced biodegradation and soil pH. However, high agricultural soil pH is often associated with greater calcium concentration through liming (adding calcium carbonate) soils with pH below agronomically desirable levels. We have found that high soil pH alone is insufficient to exacerbate enhanced biodegradation. Rather, a powerful synergy is established when high levels of calcium occur in the same soil. Reassessment of soil pH targets and adjustment methods less reliant on calcium-rich alkalis offer scope to reduce the risk of enhanced biodegradation.

Ethanedinitrile: a promising replacement soil fumigant for methyl bromide

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Methyl bromide, a long-used powerful broad-spectrum soil fumigant, is scheduled for withawal from soil use in 2005 under the Montreal protocol. Many highly intensive horticultural industries with extreme market-iven or mandated sanitation demands face significant challenges in maintaining equivalent plant protection efficacy with alternatives. Most current alternatives have a narrower activity spectrum, greater risk of phytotoxicity entailing economic penalty in longer plant-back times, are susceptible to enhanced biodegradation (a concept alien to the methyl bromide experience) or are generally less forgiving for robust use in the highly diverse and variable field environment. CSIRO Entomology has identified and taken patent protection on ethanedinitrile (C_2N_2) (EDN) as a potential soil fumigant. Its chemical properties place it at no risk as an ozone-depletor or greenhouse gas and it is safer to use than methyl bromide. Laboratory and preliminary field tests have shown that it offers equal, if not greater, spectrum of activity and potency than methyl bromide, and an extraordinarily rapid action and rate of dissipation that promises extremely short plant-back times and little prospect that enhanced biodegradation will impede efficacy with longer-term use.

Insect resistance traits in the near future

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Since their introduction nearly 10 years ago, insect resistance (IR) traits have become a significant feature of the crop protection landscape. In spite of the rapid adoption of this technology, there are relatively few IR traits currently in the marketplace, and they are only available in two major crops (maize and cotton). A limited number of new IR traits are in the product development pipelines of the major crop protection companies with projected product launches within the next 5 years. What is known about these new IR traits will be discussed. The discovery and development process for IR traits will also be discussed in the context of identifying barriers to the introduction of new IR traits.

Efficacy of a spinosad suspension concentrate formulation for controlling chewing and sucking lice on cattle under field conditions

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Field studies were conducted with cattle in Wisconsin and Illinois between January and April, 2000, to determine the efficacy of 0.04% spray and 2 mg/kg pour-on treatments of a spinosad suspension concentrate formulation against chewing and sucking lice. Positive control substances included in the study were Co-Ral® EC (5.8% coumaphos) and CyLence™ Pour-on (1% cyfluthrin). Animals in the negative control group remained untreated. All lice species were visually sampled on each animal at weekly intervals after treatment, for eight weeks. Individual louse counts were summed over each examination site for each species. A repeated measures analysis using a mixed model (SAS PROC MIXED), was conducted for data pooled across both study sites. Both spinosad treatments and CyLence™ Pour-on provided excellent efficacy (98-100%) against *Bovicola bovis* over the entire study period; Co-Ral® EC efficacy dropped below 90% from Weeks 6-8. The efficacy of the spinosad spray treatment against *Linognathus vituli* was >93% for the entire study and >88% for the spinosad pour-on treatment. Co-Ral® EC was >90% efficacious against *L. vituli* during Weeks 1-7, while CyLence™ Pour-on only exceeded 90% for Weeks 3-7. *Solenopotes capillatus* were found only at the Wisconsin site; all treatments were highly efficacious throughout the study period.

Progress of *Bombyx* genome project in Japan

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In Lepidoptera, the genome information had been quite limited so far. Now the situation changed quickly, since National Institute of Agrobiological Sciences (NIAS) is performing the *Bombyx* genome project including *Bombyx* whole genome shotgun sequencing, cDNA collection and construction of physical map systematically in collaboration with many groups, and the explosive accumulation of *Bombyx* genome information will be expected. We will present the recent progress on our *Bombyx* genome works.
a)EST db: Recently, *Bombyx* EST db, "SilkBase*"*, was transferred to "KAIKObase" http://sgp.dna.affrc.go.jp,, where EST db is combined with genome and map information as the integrated database.
b)Linkage map: About 2,000 genetic markers were mapped in 28 linkage groups under the efforts of several groups. The high density map was made public as "BombMap" in KAIKObase.
c) BAC library: Three BAC libraries prepared by different restriction enzymes are available.
d) EST microarray: EST microarray containing 6,000 non-redundant ESTs was available for functional studies.
e) Genomic sequence: NIAS carried out *Bombyx* WGS, whose data will be public in KAIKObase of NIAS HP shortly.

Reduced sensitivity of acetylcholinesterase in green rice leafhopper, *Nephotettix Cincticeps* Uhler, resistant to organophosphorus and *N*-methyl carbamate insecticides

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Green rice leafhopper (GRLH) showed resistance to various organophosphorus and *N*-methyl carbamate insecticides. Major resistance mechanism is a reduced sensitivity of acetylcholinesterase (AChE) to these insecticides. A hydophilic form of AChE was purified from *N*-methyl carbamate susceptible (SA) and highly *N*-methyl carbamate-resistant (N3D) strains of GRLH. Both of purified AChE from SA and N3D strains displayed the highest activities towards acetylthiocholine (ATCh) at pH 8.5. In the SA strain, the optimum concentration for ATCh, propionylthiocholine (PTCh), and butyrylthiocholine (BTCh) were about $1x10^{-3}$, $2.5x10^{-3}$, and $1x10^{-3}$ M, respectively. However, in the N3D strain, substrate inhibition was not identified for ATCh, PTCh, and BTCh to $1x10^{-2}$ M. The V*max* value in the N3D strain was 1.75- and 3.15-fold higher for ATCh and BTCh than that in the N3D strain. However, it was 1.13-fold lower for PTCh. The increased activity of AChE in the N3D strain is due to the qualitatively modified enzyme with a higher catalectic efficiency. AChE from the N3D strain was 65-fold less sensitive than SA strain to inhibition by propoxur. In contrast, AChE from the N3D strain 65-fold more sensitive to inhibition by monocrotophos than AChE from the SA strain. This indicated negatively correlated cross-insensitivity of AChE to propoxur and monocrotophos.

Large scale field trials in the USA with Bt cotton

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Field studies were conducted in 2000-2002 to compare arthropod populations between Bollgard$^{\text{Ò}}$ and conventional cotton season-long in Alabama, Georgia, and South Carolina. For each region, three or four paired fields were evaluated weekly, with each pair consisting of a Bollgard$^{\text{Ò}}$ field and a conventional cotton field (treated for lepidoptera when necessary). Insect species sampled included *Heliothis virescens*, *Helicoverpa zea*, *Spodoptera frugiperda*, *Spodoptera exigua*, *Trichoplusia ni*, *Pseudoplusia includens*, phytophagous stink bugs and plant bugs, cotton aphids, *Solenopsis invicta*, *Geocoris* spp., *Orius* spp., spiders, parasitic wasps, green and brown lacewings, and *Nabis* spp. Most results show that there were no significant differences for any arthropod population between a particular Bollgard$^{\text{Ò}}$ field and the corresponding conventional cotton field. However, the most amatic significant differences occurred in South Carolina when comparing conventional cotton sprayed with at least several applications of insecticides to control primarily *H. zea*, and Bollgard$^{\text{Ò}}$that was not treated for *H. zea*. In these instances, there were significantly more non-target arthropods in the Bollgard fields than in the conventional fields. These results demonstrate that the use of Bollgard$^{\text{Ò}}$usually has no impact on non-target arthropod populations, and when compared with cotton treated with insecticides for lepidoptera, results in significantly more non-target arthropods.

Molecular diagnosis of *para*-locus mutations in head lice (*Pediculus humanus capitis*) from around the world: their role in resistance to synthetic pyrethroids?

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Throughout the world head lice (*Pediculus humanus capitis*) infest people, and the pediculicides used to treat them do not always work. One group of chemicals applied to treat head lice are the synthetic pyrethroids, and resistance to these chemicals has been detected in lice in many parts of the world using the World Health Organisation (WHO) mortality assay. The genetic basis for resistance in lice to synthetic pyrethroids is hypothesised to be due to mutation in the *para*-orthologous sodium channel α-subunit (*para*-locus); where most of the sodium channel gene mutations that are known to be associated with knockdown resistance in insects are located. A number of point mutations have been detected in the *para*-locus of synthetic pyrethroid-resistant lice. We sequenced a region of the *para*-locus where two of these mutations have been previously found, in lice from Australia, Argentina, France, Guatemala, Thailand, Panama and the U.S. We observed the occurrence of these and other mutations and the correlation between genetic data and WHO mortality assays for many individual lice. We also developed a possible genetic test for resistance, a polymerase chain reaction restriction fragment length polymorphism (PCR-RFLP) assay to quickly and accurately diagnose individual lice as heterozygous or homozygous for genetic change to the *para*-locus.

Using genomic tools to isolate and characterise olfactory genes from the tortricid pest, *Epiphyas postvittana*

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With completion of the *Drosophila* genome, efforts have turned to insects of major economic interest for whole genome sequencing. However in the short-term whole genome sequencing will still remain beyond the reach of many projects working on individual insect species. Tools such as EST sequencing and proteomics can contribute to finding genes/proteins involved in targeted processes and create useful information for comparative genomic analysis. We are using these approaches to identify genes involved in the detection of odorants, particularly sex pheromones, from the horticultural tortricid pest, *Epiphyas postvittana*. Potential odorant binding proteins, odorant receptors and odorant hydolases have been isolated from ESTs derived from male antennae. Comparison of male and female antennal proteins on 1-D and 2-D gels reveals proteins both up and down-regulated in males relative to females, and mass spectrometry reveals some of these proteins correspond to genes from the EST collection. These techniques have given us quick access to potential olfaction genes. Functional studies of these genes to examine expression in antennae, protein structure and function, and involvement in olfaction using expression in insect cell lines and RNAi in whole insects will be described.

Expressed sequence tags from the brown planthopper, *Nilaparvata lugens* and its tissue-specific genes

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A total of about 35,000 expressed sequence tags (ESTs) were produced from various tissues and stages of *Nilaparvata lugens*, one of the serious pests of rice plant. cDNA libraries were made from two stages of eggs, fourth instar nymphs, two stages of adult ovary, testis, male midgut, female midgut, female salivary glands, female head, female thorax, female abdomen, male head and thorax, and male abdomen. There were more than 8,000 non-redundant ESTs, potentially representing unique genes. Eleven ESTs were found only from the female abdomen and ovary, and five out of the eleven were expressed only in the mature ovary. At least ten ESTs were testis-specific. Nucleotide sequences of truncated region of these cDNAs were determined by 5' and 3' RACE. BLAST search disclosed that the sequences of many gonad-specific genes were dissimilar to those in the databases. The ESTs were also used for determining whole mitochondria genome sequence, because the mitochondrial gene sequences were contaminated in the EST data. *Nilaparvata lugens* mitochondria genome size was 17,251 bp.

Prospects and potential pitfalls associated with the use of transgenic corn for fall armyworm management

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Transgenic corns that produce Bt toxins have been available for control of European corn borer in the Midwestern USA for several years. While the Bt-event in sweet corn is well expressed and also provides essentially 100% control of fall armyworms, the event in field corns provides only 60 to 85% control of this migratory, polyphagous pest. Despite the reduced potential for fall armyworm control, these transgenic products provide substantial yield gains over traditional field corns for grain and silage. With the recent development of sub-tropically adapted, Bt-enhanced field corns, acreage of Bt-corn grown for silage has spread south into the insect's overwintering region of Florida. IRM stewardship agreements standardized and enforced by the EPA to limit resistance development encourage growers to plant a proportion of their acreage to non-Bt corn reservoirs dependent upon their location within the U.S. cotton growing belts. Corn growers are increasingly pressed to produce high yields with low inputs to compete in the marketplace and are pushing to reduce or eliminate the reservoirs. These and other issues, including non-compliance and season-long availability of Bt-enhanced corn and cotton, will be examined for their potential for selection of Bt-resistance and its area wide implications for fall armyworm control.

Cost economics of Bt cotton cultivation in irrigated ecosystem

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Bollworms, *Helicoverpa armigera* (Hubner), *Earias vittella* (F.) and *Pectinophora gossypiella* (Saunders) have made cotton cultivation uneconomical. Transgenic cotton hybrids with in built δ endotoxin reduces pest problem and cost of cultivation. Studies were undertaken at Regional Agricultural Research Station (RARS) and in farmers' field. MECH-162 Bt and non Bt during 2002-03 and MECH-184 Bt and non Bt during 2003-04 and local hybrid (NHH-44) were cultivated. Sprays were imposed as and when the pest population/damage crossed ETL. Totally 3, 4 and 4 sprays during 2002-03 and 3, 12 and 12 sprays during 2003-04 were imposed on Bt, Non Bt and NHH-44 hybrids respectively in RARS, whereas, in farmers' field 4, 9, and 9 sprays during 2002-03 and 6, 12 and 12 sprays during 2003-04 were imposed on Bt, Non Bt and NHH-44 hybrids respectively. Incidence of bollworms only in Bt cotton MECH-162 and 184 was much below ETL. More good opened bolls and less bad opened bolls per plant were recorded with significantly higher seed cotton yield in Bt cotton as compared to non Bt and local hybrid. Bt cotton hybrids recorded 35 to 45 percent increase in net it over local hybrid making it commercially viable and itable.

Wound-inducible expression in transgenic plants after insect attack

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Promoters of genes that are induced by microbial pathogens, wounding and treatment with wound-signalling compound could be valuable in regulating the expression of resistance genes when plants are attacked by insects or pathogens. A plant expression vector pMP4 was developed using a GUS/GFP fusion construct driven by a *Shpx6b* peroxidase gene promoter (J. Manners, UQ) from a tropical forage legume *Stylosanthes humilis*. This construct was introduced into insect susceptible tobacco using *Agrobacterium tumefaciens*. Expression of *gus* and *gfp* reporter genes in T_1 transgenic tobacco leaves after infestation/treatment with chewing insects, sucking insects, the plant defense regulator MeJa, and mechanical wounding were investigated. *Gus* reporter gene levels were also assayed to quantify expression and the results were consistent with the observed histological patterns of expression. The results show that the *Shpx6b* promoter switches on the expression of linked genes after damage by insect herbivores, and could be useful in regulating the expression of heterologous genes for insect and /or pathogen resistance in transgenic plants.

Appraisal of cross resistance potential to neonicotinoid insecticides by *Bemisia tabaci*

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Resistance that occurs to a particular insecticide is often assumed to extend to all members of the class. However, structural differences among compounds may affect vulnerability to degradation by metabolic enzymes, or differences in target site receptors may influence susceptibility to specific insecticides, thereby mitigating cross-resistance effects. Neonicotinoids are an important class of insecticides used worldwide against *B. tabaci* and many other insect pests. Populations from southern Spain resistant to imidacloprid have also shown cross-resistance to thiamethoxam and acetamiprid. To further assess the expression of cross-resistance to neonicotinoid insecticides, *B. tabaci* populations from California and Arizona were monitored periodically to determine their susceptibilities to acetamiprid, dinotefuran, imidacloprid, and thiamethoxam. In addition, two imidacloprid-resistant strains were tested for cross-resistance potential. Field strains expressing different levels of resistance to imidacloprid did not always show cross-resistance to other compounds. For example, a population from Arizona collected in November 2003 had an LC_{50} of 115ppm to imidacloprid in a leaf-uptake bioassay, but only 8 and 9 to dinotefuran and thiamethoxam, respectively. The resistance factor was even greater in a strain from Guatemala that yielded an LC_{50} of 653ppm to imidacloprid, but only 29 to dinotefuran. Implications for cross-resistance between neonicotinoids among the various strains are examined.

Developing protocols for field evaluations of non-target organisms in Bt crops

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Transgenic crops containing insecticidal genes from Bacillus thuringiensis (Bt) bacteria offer unprecedented power to combat agricultural pests, but concerns about their possible unintended effects remain. Laboratory research suggests accidental exposure or indirect effects of Bt toxins may negatively impact non-target species, though this result is rarely seen in field studies. However, such field research sometimes employs methods inappropriate for evaluating Bt crops, which may have relatively small effects on a select number of species. Nonetheless, simple modifications to experimental protocols and analysis could greatly increase the value of field tests of non-target effects. A synthesis of existing data may guide choices of appropriate indicator taxa, needed replication, and optimal sample timing. Current statistical methods may also allow for more powerful analysis of time-series community data. Field experiments assessing the effects of plot size on community and species-level tests of non-target effects may allow for research plots which are both practical and realistic. Overall, such changes will improve researchers' power to detect differences, positive or negative, between Bt crop production and alternate management practices. However, methods developed for current Bt corn varieties should be adaptable to studies of new cultivars, non-Bt management practices, and other cropping systems.

Studies on detection and characterization of organophosphate-resistance in *Anopheles culicifacies* sibling species in Madhya Pradesh, Central India

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Field studies carried out in villages in two blocks in Chhindwara district indicated presence of malathion resistance in *Anopheles culicifacies,* the principal vector of malaria in this area, in the absence of use of malathion spraying for malaria control. *An. culicifacies* s.l. was found 36-86% resistant to malathion but was susceptible to other organophosphate, carbamate and synthetic pyrethroid insecticides. Selection of malathion-resistance in *An. culicifacies* appears to be due to insecticides used in plant protection. Biochemical assays indicated non involvement of general esterases and insensitive acetylcholinesterase in conferring malathion-resistance. Synergist-malathion bioassays indicated the involvement of specific malathion carboxylesterase (MCE). ASPCR assays have revealed prevalence of species B (73%) and species C (27%) and exhibited diferential susceptibility to malathion. The observed percent susceptibility to malathion in species B was 68% while in species C it was 13% These results will be presented in detail and the implication of development of resistance owing to selection by agricultural pesticides will be discussed.

Sugarcane borer (Lepidoptera: Crambidae) resistance management studies in Louisiana

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During the last decade, sugarcane borer (SCB), *Diatraea saccharalis* (F.) management in Louisiana has been impacted by cultural practice changes emphasizing early planting initiated in August, and the increasing dominance of a high yielding borer-susceptible variety (LCP 85-384) now approaching 88% of the planted acreage. Insecticide foliar sprays for SCB control rose from 0.2 applications per season in 1994-95 to 1.8 applications per field in 2002-03. Due to the growing popularity of the ecdysone agonist, tebufenozide (Confirm®), insecticide resistance monitoring efforts were begun throughout the sugarcane industry in 2002. Increases in LC_{50} values from traditionally low insecticide use areas have ranged up to 2.6 fold in areas averaging more than one application per season. In one area of heavy tebufenozide use, LC_{50} values remained high despite discontinuance of selection pressure. Resistance management studies will be discussed in relation to broader SCB management issues.

Twenty years of insecticide resistance management in the Australian cotton industry.

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The Australian Cotton industry has been guided by an Insecticide Resistance Management Strategy (IRMS) for twenty years. Since its introduction in the 1983/84 cotton season to combat widespread pyrethroid resistance by the primary pest of cotton *Helicoverpa armigera*, it has been modified and expanded to encompass an increased number of chemical groups, embrace integrated pest management and account for a wider range of insect pests. Compliance to this voluntary strategy is near to 100 % by the cotton industry, and in recent years the grains industry has become involved in the strategy providing a more co-ordinated attempt across multiple farming practices to manage resistance and maintain the chemicals available for pest control. This paper shall provide an overview of the IRMS, its evolution over the last twenty years, the principles involved and why uptake by the industry has been so successful. The current status of insecticide resistance by the primary pests of cotton shall be discussed, as well as the future of the IRMS, including the possible impact of transgenic cotton on the pest complex and insecticide resistance management.

Global status of insecticide resistance mechanisms in *Helicoverpa armigera*

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The cotton bollworm *Helicoverpa armigera* (Hübner) has displayed an enormous potential to resist almost all the insecticide molecules used against it thus far. Amongst the range of mechanisms that confer resistance, enhanced metabolic detoxification has been found to be the most important. Recent evidence showed that increased oxidative breakdown of pyrethroids was found to be associated with pyrethroid resistant strains from West Africa, India, China, and Pakistan. Thus far, at least four cytochrome p450 genes CYP6B2, CYP6B6, CYP6B7 and CYP4G8 have been implicated in pyrethroid resistance. Pyrethroid resistance due to enhanced esterases, reduced penetration and nerve insensitivity was reported from India, China, Pakistan, Thailand and Australia. Several genes associated with nerve-insensitive resistance were explored for specific structural changes, but the mutations generally associated with sodium channels and GABA chloride channels in other insecticide resistant species were not found in *H. armigera* and those that were reported previously in the intracellular segment linking repeat domains III and IV of the para-homologous sodium channel gene, that were associated with nerve insensitive *H. armigera* larvae, were not confirmed. Insensitive acetylcholinesterase and or unique esterase isozymes were found to be associated with resistance to organophosphates and carbamates. Resistance diagnostic kits based on the unique resistance associated isozymes, have been made in India.

Resistance issues in Bt cotton in India and China

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Genes for resistance to Cry toxins being selected in the field *in Helicoverpa armigera* by Bt cottons may not be necessarily those which are also being found in laboratory selections. Resistant individuals are present in the field at frequencies of between one in $0.42x10^{-3}$ and one in $1.3x10^{-3}$ in India and China. There appears to be both a recessive target site mechanism and a dominant enzymatic degradation mechanism. Currently the former seems likely to be more important in Chinese field populations and the latter in Indian populations with important implications for resistance management strategies. A simulation model was produced taking account of the biology of the pest, the local cropping pattern, initial resistance gene frequency, dominance, mode of inheritance, relative survival rates of RR, RS and SS individuals on Bt and non-Bt hosts throughout the year and other variables. Factors of importance in determining the time to a filed resistance gene frequency of 0.5 in the two countries are different. Where resistance is semi-dominant, in-crop refugia were of little value in delaying resistance development. However even fitness costs as low as 5% doubled the time to significant resistance in most scenarios. Integrating other artificial mortality factors at times of peak pest presence was the simplest effective strategy for delaying resistance in systems where >40% of the cotton is Bt transgenic. The likely impact on resistance development of the range of dual gene combinations currently being commercialised in the two countries has been modeled and is discussed.

Survival and development of *Spodoptera frugiperda* and two of its natural enemies on Bt transgenic maize

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This paper reports effects of conventionally-bred partially resistant and transgenic maize hybrids on *Spodoptera frugiperda* (herbivore), *Campoletis sonorensis* (parasitoid) and *Beauveria bassiana* (entomopathogenic fungus). *Spodoptera frugiperda* larvae fed maize expressing the Bt toxin Cry1Ab had an increased development time and reduced pupal weights compared to larvae fed conventionally-bred resistant and susceptible maize hybrids. *Campoletis sonorensis* cocoon and adult weights were significantly reduced by 15-30% when reared in hosts fed Bt maize. The extent of this effect varied with the weight of host at oviposition, its subsequent development and host weight at parasitoid emergence. Resistant maize, whether transgenic or conventionally-bred, may increase the susceptibility of *S. frugiperda* to *B. bassiana*, as larvae develop more slowly than on susceptible maize, with greater time intervals between moults. This increases the time for *B. bassiana* spores to germinate and invade the host through the cuticle before they are shed during moulting. Alternatively, resistant plants may induce an immune response in *S. frugiperda*, reducing susceptibility to fungal attack. Results of ongoing experiments on this interaction will be presented.

The *Diabrotica* genetics consortium: a paradigm for global cooperation in research

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The evolution and spread of insecticide- and rotation resistance, the introduction and spread of the western corn rootworm (*Diabrotica virgifera virgifera*) in Europe, and the recent large-scale commercial deployment of rootworm-active *Bt* corn in North America have generated a growing sense of urgency among scientists involved in rootworm genetics research. These are all complex problems, with a number of researchers in several countries working on various aspects. In the last year, a group of more than 35 scientists from 25 different institutions in the U.S., Canada, France, and Germany have come together to form the *Diabrotica* Genetics Consortium. The main role of the Consortium is to facilitate communication among those working on related problems, both to avoid unnecessary overlap of effort and to reveal opportunities for synergism through cooperation/collaboration. It is founded on the principle that open communication of research goals, activities, findings, and future plans is in the best interest of all concerned and in the best interest of advancing science. The scientists comprising the Consortium are committed to sharing information and ideas in an atmosphere of mutual trust, respect, and sensitivity to the essional needs of one another in accord with the highest ideals of scientific endeavor.

Induction and transmission of Bt-tolerance in the flour moth *Ephestia kuehniella*

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The use of *Bacillus thuringiensis* endotoxins to control insect vectors of human diseases and agricultural pests is under threat from the possible evolution of resistance in major pest species. In addition to high levels of resistance, produced by receptor insensitivity, several cases of tolerance to low to medium levels of toxin have been reported in laboratory colonies of lepidopteran species. Since the molecular basis of some of these cases of tolerance to the toxin are not known, we explored alternative mechanisms. Here we present evidence that tolerance to a Bt-formulation in a laboratory colony of the flour moth *Ephestia kuehniella* can be induced by pre-exposure to a low concentration of the Bt-formulation and that the tolerance correlates with an elevated immune response. The data also indicate that both immune induction and Bt-tolerance can be transmitted to offspring by a maternal effect and their magnitudes are determined by more than one gene.

Effects of reduced prey choice caused by GM plants on a predatory insect

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Bt plants are highly toxic to certain pest species. Predatory insects may be affected by the reduced abundance of target pests if sufficient alternative prey is not available. This paper will present experiments investigating such indirect effects under laboratory conditions. Lacewing (*Chrysoperla carnea*) larvae were released into large cages containing Bt and wildtype plants previously infested with a constant number of diamondback moth (*Plutella xylostella*) eggs but different numbers of aphids (*Myzus persicae*). No *P. xylostella* larvae survived on Bt plants but aphid populations reached high densities on both plant types. Lacewings recovered from Bt plants at the end of the experiment did not differ in weight from ones recovered from wildtype plants. At the highest initial aphid densities there was also no difference in the proportion of lacewings surviving on the two plant types. Significantly fewer lacewings were recovered from Bt plants than from wildtype plants as initial aphid numbers were reduced. Interestingly, however, numbers of *C. carnea* recovered from wildtype plants were only significantly correlated with numbers of surviving *P. xylostella* at the highest aphid densities. Possible roles of escape behaviour of *P. xylostella* as well as cannibalism between *C. carnea* larvae in these interactions will be discussed.

The effect of spinosad on different developmental stages of Colorado potato beetle *Leptinotarsa decemlineata* (Say) (Col.: Chrysomelidae)

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The Colorado potato beetle (CPB) *Leptinotarsa decemlineata* is a major insect pest of potatoes. In this study we investigated the effect of spinosad on egg, larval and adult stages of CPB. A strain of CPB was collected from Ardabil, where this pest was reported for the first time from Iran in 1984; and was reared in greenhouse conditions. Bioassays were conducted using eggs, neonates, 1st, 2nd, 3rd and 4th instar larvae as well as adults. In preliminary experiments, no ovicidal effect was observed. The concentrations used for treating the larval stages and adults ranged from 1-80 ppm. To estimate the LC values, each treatment was replicated 4 times. Bioassays were conducted at 26 ± 1 °C and 16:8 (L:D) h photoperiod in transparent plastic containers. The mortalities were recorded 24 hrs after treatment. Based on probit analysis of the data, LC_{50}s were 2.1, 3.2, 4.6, 6.5, 20.2 and 12.0 ppm and the estimates for LC_{90} values were 8.0, 12.9, 17.0, 30.1, 67.8 and 30.2 ppm for neonates, 1st, 2nd, 3rd and 4th instar larvae and the adults, respectively. If spinosad also performs well in the field, this insecticide will be a suitable candidate for CPB management programs in the region.

Laboratory selection for resistance to Bt toxins in the European corn borer: potential and limitations

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Transgenic maize hybrids expressing toxins from *Bacillus thuringiensis* have been available for managing European corn borer (*Ostrinia nubilalis* Hübner) populations since 1996. However, there is concern that this technology will result in widespread resistance to Bt toxins. The "high dose-refuge strategy" is the currently accepted method for managing resistance in *O. nubilalis* and has become a requirement of registration for all transgenic events. However, this strategy is based on a number of assumptions that are difficult to validate until resistance is identified and characterized. Resistant strains resulting form laboratory selections provide a potential tool to assess these assumptions in the absence of field derived resistant populations. However, such laboratory experiments are limited by their inability to reflect the selection intensity imposed on a field population exposed to transgenic plants. Our laboratory has been selecting *O. nubilalis* by exposure to Bt toxins incorporated into an artificial diet since 1998. A number of different colonies that vary with regard to the nature of the resistance, degree of dominance, number of genes and ability to survive on transgenic plants have been developed. The relevance of these strains in terms of their ability to improve and refine resistance management recommendations will be discussed.

Insecticide mode of action classification: a key to insecticide resistance management

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The Insecticide Resistance Action Committee (IRAC) has developed a classification scheme for insecticides (and acaricides) that is based on mode of action (MoA). Experience has shown that effective insecticide resistance management (IRM) strategies seek to minimise the selection for resistance for any one type of insecticide or acaracide. In practice, alternations, sequences or rotations of compounds from different MoA groups provide a sustainable and effective approach to IRM. This ensures that selection for resistance through the use of compounds in the same MoA group is minimised. It is recognized that insecticide resistance can also result from enhanced metabolism by enzymes within the pest, reduced penetration of the pesticide into the pest or behavioural changes of the pest. These resistance mechanisms are not linked to any site of action classification, but are specific for individual compounds or chemical groupings. Despite this, alternation of compounds from different chemical classes remains an entirely viable resistance management technique because such a practice will always minimise selection pressures. This presentation describes the mode of action classification scheme and its use in IRM.

Insecticide resistance surveillance of disease vectors in Peru

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Vector-borne diseases, such as malaria and dengue, are significant public health problems in many countries of South America, including Peru. While environmental modifications and infrastructure improvements to reduce vector populations are the ideal, the reality in many areas of the world is that insecticides play a crucial role in vector suppression. Resistance to these insecticides can potentially interfere with vector control operations. In Peru, there is little information concerning insecticide resistance in *Anopheles darlingi* or *Aedes aegypti*, the principal vectors of malaria and dengue, respectively. Utilizing the CDC bottle bioassay, personnel from the U.S. Naval Medical Research Center Detachment, Lima and the Peruvian Instituto Nacional de Salud, have conducted over 3,300 assays of over 70,000 *An. darlingi* and *Ae. aegypti* mosquitoes to establish diagnostic concentrations for 8 insecticides representing 3 chemical classes. In addition to the assays, a workshop was held to train 22 technicians from 11 regional reference laboratories in Peru on the CDC bottle bioassay methodology. The goal of these assays and training is to ultimately establish a national insecticide resistance monitoring program and assist the Peruvian health authorities in implementing effective vector control programs.

Mechanisms underlying mite outbreaks following imidacloprid applications: effects on mite fecundity and natural enemy communities in landscapes.

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Imidacloprid, a chloronicotinoid insecticide has become one of the most widely used products for insect control. It provides excellent control for many pests; however, following the elimination of the primary pest, secondary infestations of mites have been observed. The mechanisms underlying this phenomenon are thought to include alterations in mite fecundity levels stimulated by sublethal levels of the insecticide (hormoligosis), and disruption of natural enemy communities. To investigate the effect of imidacloprid on beneficial insects, we examined the influence of the insecticide on longevity and survival of a general predator, *Orius insidiosus*, and abundance and diversity of natural enemies on imidacloprid treated plants. Hormoligosis in boxwood mite (*Eurytetranychus buxi*) was also investigated. *Orius insidiosus* was shown to be negatively affected by imidacloprid in plants. However, the study of insect diversity and abundance indicated that beneficial insect communities differed only slightly between imidacloprid treated and untreated sites. Moreover, we were able to create increased densities of mites and attendant damage in the absence of predators. The results of the study suggest that the mechanisms leading to increased mite populations involve several aspects of mite and natural enemy ecology and, possibly, changes in mite or plant physiology.

Delayed resistance to Bt crops: is Arizona's story unique or universal?

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Transgenic cotton producing *Bacillus thuringiensis* (Bt) toxin Cry1Ac that kills pink bollworm, *Pectinophora gossypiella*, has accounted for more than half of Arizona's cotton since 1997. Despite the wide use of Bt cotton and surprisingly high frequency of pink bollworm resistance to Cry1Ac detected in 1997, Bt cotton has remained extremely effective against pink bollworm. Laboratory bioassays show that the frequency of resistance to Cry1Ac did not increase from 1997 to 2003. Field-based estimates also show sustained efficacy during this period. In laboratory-selected strains with up to 3,100-fold resistance to Cry1Ac and survival on Bt cotton plants, resistance is linked with three recessive mutations in the gene encoding a cadherin protein that binds Cry1Ac. DNA-based monitoring of field-sampled individuals using the polymerase chain reaction (PCR) indicates that the frequency of the three identified resistance alleles remains low. Other examples of delayed resistance to Bt crops have been reported from Australia, China, and elsewhere in the USA. Factors delaying pest resistance to Bt crops include refuges of non-Bt crops, low initial resistance allele frequency, recessively inherited resistance, fitness costs, and incomplete resistance. The success of Bt crops exceeds expectations of many, but does not preclude future resistance problems.

Construction of transgenic *Bombyx white egg-1* mutant introduced kynurenine 3-monooxygenase gene

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Bombyx white egg 1 mutant (*w-1*) is characterized by the white colour egg and eye. It was known that the mutant kynurenine 3-monooxygenase gene possesses a partial in the exons. To understand an effect of complementation of wild type *KMO* gene, we introduced *KMO* cDNA with the promoter of native *KMO* or cytoplasmic actin gene (*A3*). We obtained three transgenic lines with A3 promoter and one line with the native promoter. The adult eyes of these lines turned to be brown and the eggs laid by the transformed females were also brown. The analysis by RTPCR showed that the lines with *A3KMO* gene accumulated the transcript in the mid gut, fat body and Malpighian tubes. A line with *KMO* promoter expressed it only in the fat boy and Malpighian tubes. The strength of eye and egg colour in the transgenic line was proportional to the *KMO* expression level. Interestingly, the transgenic larvae with *A3KMO* gene changed the larval skin colour more brownish. These results suggest that the morphological change in *w-1* mutant is caused by a deletion of *KMO* gene. The mutant and *KMO* gene can be used as a host and marker for a construction of transgenic silkworm.

Presence of avidin and aprotinin in the bodies and frass of *Spodoptera litura* (Lepidoptera: Noctuidae) larvae feeding on transgenic tobacco plants.

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To determine whether herbivorous insects and their frass could be environmental sources of novel proteins from transgenic plants, *Spodoptera litura* larvae were fed on transgenic tobacco leaves expressing either the biotin-binding protein, avidin, or the protease inhibitor, aprotinin. Control larvae received non-transgenic tobacco. Samples of 20 larvae each were taken on Days 5, 6 and 7 and two samples of frass on Days 6 and 7. Insects in all treatments grew significantly between Days 5 and 7, but the avidin-fed larvae were significantly smaller and produced less frass than the others on Day 7. All insects produced more frass on Day 7 than on Day 6, but the mass of frass produced per mg insect did not vary from day to day. Both novel proteins were present in larvae at lower concentrations than in the transgenic leaves they had fed upon. Unbound avidin was found in the samples of avidin-fed larvae and their frass. Aprotinin was detected in all larval and frass samples from insects fed aprotinin-tobacco. Unbound aprotinin accounted for only a portion of that found in larvae and in frass.

Bollgard® II cotton, tested for the first time in West Africa, shows excellent lepidopteran control and potential for improved cotton production

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Field studies were conducted in Burkina Faso during the 2003 cropping season to evaluate the efficacy, against lepidopteran insects, of cotton expressing two endotoxins of *Bacillus thuringiensis* subsp. *kurstaki* (Berliner), commonly known as Bollgard® II cotton. Bollgard® II, treated against sucking and piercing pests only, was compared, in two research stations, to local varieties treated against lepidopteran insects and sucking pests. Our data showed that Bollgard® II cotton provided control of major lepidopteran species that attack cotton in Burkina Faso. The average number of bollworm larvae per plant over time was significantly different between transgenic cotton (treated against sucking pests only) and a local variety (treated against lepidopteran insects and sucking pests). The average number of *Helicoverpa armigera* larvae per Bollgard® II plant was reduced by 92% when compared to the local varieties. Bollgard II and the local spraying regimen were similarly highly effective in reducing foliar feeding lepidopterans. The difference in damage by lepidopteran insects, resulted in fewer abscissions of floral and fruiting bodies and higher numbers of bolls per plot in the Bollgard® II cotton. In Frako-Bâ and Kouaré evaluation sites, Bollgard® II cotton rendered a yield 13% and 19% higher, respectively, than the commercial local variety.

Bt crops in Argentina: efficacy and resistance management

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Bt crops began to be de-regulated in 1998 (Bt Cotton: MON 531; Bt Corn: E 176, and MON 810). In 2001, Bt 11 was also authorized. Both Bt crops have steadily increased their acreages. Last season, 45 % of the corn area was planted with Bt corn, and 20% of the cotton was Bt. Bt corn produces Cry1Ab protein, and Bt cotton produces Cry1Ac protein. Corn and cotton producing areas do not significantly overlap, and the only lepidopteran pest in common for both crops in the country is *Spodoptera frugiperda.* Target pests for Bt-cotton are: *Alabama argillacea, Heliothis virescens/Helicoverpa gelotopoeon* complex, and *Pectinophora gossypiella.* For Bt-corn, the target species is *Diatraea saccharalis.* The Insect Resistant Management (IRM) group proposes strategies for the management of Bt crops, based on local and external experiences. Research is conducted by third party entomologists from Universities, and National Research Institutes. Mainly, the the strategy is based on a required refuge (non-Bt crop), which is 20% for Bt-cotton, with the possibility of insecticidal-non-Bt applications as needed, according to economic thresholds. For Bt-corn, the requirement is 10% refuge without any insecticides for the target pest. IRM strategy also considers studies such as baselines, emergency plans, monitoring, and continuous studies related to the size of the refuge, that will be discussed during the presentation.

Altered gene expression levels linked to fenvalerate resistance in cotton bollworm, *Helicoverpa armigera*

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The ANO2 strain of *Helicoverpa armigera* is 50-fold resistant to the pyrethroid insecticide fenvalerate; largely due to a single gene, *RFen1*, on Linkage Group 13. The cDNA-AFLP technique was used to search for changes in gene expression levels controlled by or linked to *RFen1*. Larvae from a segregating backcross family were genotyped at a linked marker locus, enabling prediction of their fenvalerate resistance status without exposure to insecticide. mRNA was isolated from individual larvae and used to make cDNA, which was processed for AFLPs (double restriction digestion, ligation to adapters, amplification by two rounds of PCR, and resolution of labelled fragments on a sequencing gel). 256 different primer combinations were tried and a total of 426 bands showing differences between resistant and susceptible groups were identified. Three were cytochrome p450s, with similarity to *Depressaria pastinacella* CYP6AE1 and *Helicoverpa armigera* CYP4S1 and CYP4AP1. The latter was upregulated whereas the other two were down-regulated in the resistant group. In addition, two carboxylesterases were down-regulated and one was up-regulated in the resistant group. Currently, real time RT-PCR is being used to verify the results and full length sequences of these potential detoxification genes are being sought using rapid amplification of cDNA ends (RACE).

Bt: Organic production – new life for the oldest commercial biopesticide.

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The Organization for Economic Cooperation and Development predicts that biopesticide may grow to 20% of the world's pesticide market by 2020. Biopesticides represent approximately 1% of the world pesticide market, and Bt products represent > 75% of biopesticides sold. Other organizations like the World Health Organization, Food and Agriculture Organization and European Commission are also predicting dramatic growth for biopesticides over the next 10-20 years. Sprayable Bt formulations have penetrated cotton, fruit and vegetable, aquatic and other insecticide markets in the US, EU and the world. Organic producer dependence on Bt for worm and, in a more limited fashion, beetle control globally. In fact, many organic workers claim that Bt is more important to the organic community than to other producers because conventional producers have a greater array of management tools to choose from. With organic produce sales increasing steadily worldwide, Bt use and resistance management should be an important consideration in any system that anticipates frequent application, but especially in organic production systems where use can be extreme. Fortunately, the organic formulation of Spinosad™--a fermentation macrocyclic lactone product, is changing management practices in many cropping systems and this may change Bt's organic market dominance. In addition, new insect growth regulators and other novel modes of action may be entering the organic market in the next few years providing new opportunities to implement truly integrated management programs in organic production.

The international arthropod resistance database and newsletter: a report

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A number of resources related to resistance management have been cultivated at Michigan State University in the USA. This presentation will highlight these developments and how resistance workers all over the world can access and participate. The first of these is the Resistant Pest Management Newsletter (RPM News), which has a worldwide circulation of roughly 2,500 resistance workers in academia, industry, and government. It provides information on ongoing changes and advances in the field of resistance management, and is produced in cooperation with the Insecticide Resistance Action Committee (IRAC), the US Department of Agriculture (CSREES) and the US Western Regional Coordinating Committee (WRCC-60). Our goal in implementing a resistance database is to help address arthropod resistance by evolving an integrated, cooperative system to effectively deliver resistance information efficiently. Our information currently resides in an electronic database at Michigan State University's Center for Integrated Plant Systems and is available via the Internet at http://www.cips.msu.edu/resistance/rmdb/. The database design features a central table of 'resistance' records with five key pieces of information: the arthropod, the pesticide, the location (country), the year, and a reference to the document reporting the case. There are secondary tables that elaborate on these five key informational groups as well as auxiliary tables to help refine the information further or link to external data sources.

Juvenile hormone bioanalogs – selectivity, importance and perspectives in insect pest management

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Insect juvenile hormone bioanalogs (JHAs, juvenoids) have been the target of the long-term research in our team for many years. A special interest has been paid to several series of 2-(4-hydoxybenzyl)cycloalkanols for their high biological activity values obtained in screening tests to a broad variety of insect species. Even if a number of these JHAs have been proven to be stable but non-persistent under the environmental conditions, designing of ways for limited increase of their stability in environment and for modification of their physico-chemical properties had been performed. This effort resulted in designing and developing the pioneer hormonogen substances (juvenogens). The juvenogen molecule consists of a JHA, bearing convenient functionality, which can be easily transformed into another functionality by chemical means or by enzymic ways. This step results in a synthesis of a biologically inactive or low active substance, which is characterized by physico-chemical properties different from those found with the parent JHA. The new substance, a juvenogen (a hormonogen chemical species), can be prepared as a targeted species, a substance bearing physico-chemical properties designed in advance. This strategy enables to synthesize "juvenogens upon demand", which are designed for application on selected insect species under selected laboratory or environmental conditions.

UK-wide studies of biodiversity impact of GMHT crops

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By the end of 1998 four genetically modified herbicide-tolerant (GMHT) crops were nearing the end of the regulatory hurdles for commercial growing in the UK. Although human health and direct environmental effects had been assessed and they had been declared safe the question was raised about indirect environmental effects caused by more efficient weed control. The result was the Farm Scale Evaluations, one of the largest agro-environmental impact studies ever undertaken, covering plant and invertebrates at 60-70 field sites for each of 4 crops over a three year period. The background and results from these trials will be discussed in relation to wider issues about insect biodiversity in agricultural landscapes.

Current status and future prospect of Bt cotton commercialization in China

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Transgenic cotton that expressed a gene derived from the bacterium *Bacillus thuringiensis* (Bt) was introduced for combating *H. armigera* in 1997, and rapidly increased to 58% of the total cotton area of 4.8 million ha. in 2003. In the Yellow River cotton-farming region of Northern China, where *H. armigera* has historically caused major yield losses, Bt cotton has been exclusively planted. The pest management tactics associated with Bt cotton planting causes astic reduction in use of insecticides due to the high efficacy of the transgenic cultivars in suppression of *H. armigera* infestations, increases the abundance of beneficial insects and improves the natural control of some pests. The monitoring results indicate that movement toward Bt cotton resistance in *H. armigera* is not apparent in face of intensive planting of Bt cotton over several years, which could be associated with the migratory behavior of the insect and the natural refuges derived from the mixed planting system of cotton, corn, soybean and peanut on single- family holdings with a small scale under one half hectare.

The involvement of microsomal oxidases in pyrethroid resistance in *Helicoverpa armigera* from Asia

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Five contemporary strains of the bollworm *Helicoverpa armigera* Hübner from China, Pakistan and India, all with high resistance to pyrethroids, were compared with a standard susceptible strain that originated from the Cote D'Ivoire in the 1970s ('SCD'). The strong synergism of pyrethroids by PBO implied that an oxidative metabolism could be involved in pyrethroid resistance in all resistant strains. The activities of cytochrome P450 monooxygenases from midguts of final instar larvae to *p*-nitroanisole (PNOD), ethoxycoumarin (ECOD), methoxyresorufin (MROD) significantly increased in all the resistant strains when compared with the susceptible strain. This further implies that cytochrome P450 monooxygenases are involved in pyrethroid resistance in Asian *H. armigera*. Comparative *in vitro* studies of the metabolism of ^{14}C-deltamethrin by midgut microsomes of the resistant PAK and susceptible SCD strains showed that the resistant strain had a much greater capacity than the susceptible strain for the metabolic degradation of deltamethrin. This enhanced metabolic degradation occurred in the presence of NADPH which suggested an oxidative detoxification. In the resistant strains, minor increases in glutathione S-transferase activity (to the substrates CDNB and DCNB), and esterase activity (to the substrate α-naphthyl acetate) further suggested that, of the putative metabolic mechanisms, oxidases are the most important. This study provides the first evidence that cytochrome P450 monooxygenases are a major metabolic mechanism responsible for pyrethroid resistance in *H. armigera* from Asia.

Bt rice studies of ecological risk and nontarget effects in China

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Bt rice has been permitted for field trials since 1998. After then, impacts of our different Bt rice lines containing *cry*1Ab or *cry*1Ab/*cry*1Ac gene with high resistance against rice stem borers and leaffoles, on the arthropod community and population dynamics of some dominant nontarget phytophages and spiders under field conditions was evaluated using different sampling methods. The results showed that our Bt rice lines generally did not have any significant negative effects on the arthropod community in rice paddy habitat, except that sometime resulting in the marked decrease in the abundance of some parasitoids of target pests such as braconids and ichneumonids. These tested lines also did not act any adverse impacts on the predation, growth and development, reproduction of two common spiders, *Pirata subpiraticus* and *Ummeliata insecticeps,* using either leaffolders or planthoppers feeding on Bt rice plants as their prey. In contrast, some Bt rice lines might not be suitable for the brown planthopper, *Nilaparvata lugens* to feed and lay eggs. In addition, no significant adverse effects were found on the survival, growth and development as well as the fecundity of the domestic silkworm, *Bombyx mori* fed with the mulberry leaves contaminated by Bt rice pollen grains.

In vivo effect of PBO on resistance-associated esterases from Australian *Helicoverpa armigera* Hübner (Lepidoptera: Noctuidae)

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Piperonyl butoxide (PBO) is a commercially important and successful insecticide synergist against a variety of insects in house-hold situations and against public health pests. In agriculture use has been restricted to use against the cotton bollworm, *Helicoverpa armigera* (Hübner) (Lepidoptera:Noctuidae). It has been deployed in the field to help partially restore the efficacy of pyrethroids that are currently compromised by metabolic based resistance. It has been established that PBO inhibits some insect esterases associated with insecticide resistance (Gunning *et al.*, 1998; Moores *et al.*, 1998; Young *et al.*, 2002; Young *et al* 2003). Kinetic esterase assays using *H. armigera* reveal that esterase activity was inhibited by the insecticide synergist, PBO, over a 24hr period. Esterase inhibition by PBO did not occur immediately after doseage but occurs with maximum enzyme inhibition being obtained several hours later. Laboratory analysis with *H. armigera* and pyrethroids confirmed that maximum synergism occurred at the pre-treatment times corresponding to maximum *in vivo* esterase inhibition in biochemical studies. These results were confirmed in the field when using a 5 hr pre-treatment of PBO prior to pyrethroid exposure. These results indicate that with improved temporal application of PBO, it may be possible to restore some pyrethroid efficacy in *H. armigera*. It would also be expected that partial restoration of efficacy with other conventional insecticides currently compromised by esterase based resistance mechanisms would occur.

PTD significantly synergize Bt and overcome the low resistance of diamondback moth to Cry1Ac

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Protein Transduction Domains (PTD) are kinds of peptides which can automatically penetrate bio-membrane and be acted as a carrier to transfer foreign molecule into cell(Science. 1999,285(3):1569-1572.). A fusion gene,pET21b-TAT-Cry1Ac, consisting of Cry1Ac and TAT(a kind of PTD) ,as well as its control pET21b-Cry1Ac was constructed and expressed in E.coli. The toxicity of this two expressed and purified fusion proteins to Diamondback moth were determined. To the susceptible strain the LC50 of Cry1Ac and TAT-Cry1Ac are 20.55 mg/L and 4.25 mg/L; and 124.23mg/L and 12.81 mg/L to the low resistant strain. TAT-linked Cry1Ac show a synergistic ratio of 4.84 folds to the susceptible strain, 9.70 folds to the resistant strain, and restore the susceptibility of low resistant Diamondback moth to Cry1Ac. PTD show strong potential in synergizing Bt insecticidal protein and overcoming Bt resistance.

Fluctuating asymmetry of target and non-target insects in response to the stress of GM cotton

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Fitness and fluctuating asymmetry (FA) in response to the stress of GM cotton were examined among the different symmetric characters of target and non-target pests (cotton bollworm, *Helicoverpa armigera*, CB, and cotton aphid, *Aphis gossypii*) and a beneficial species (Asian lady beetle, *Harmonic axyridis*, ALB). The results indicate that non-target effects do arise from some GM cultivars. The FA values of CB in lab decreased gradually generation by generation. The light-trap catches of CB collected in northern China with different planting history of Bt-cotton (1~5 years) also exhibited a similar trend on FA. Bt-cotton and conventional cotton are equally suitable hosts for cotton aphid. The fitness and FA of ALB and cotton aphid were similarly when fed on the aphids from both Bt-cotton and conventional cotton. But the Bt+CpTI cotton present significant impacts on the fitness of cotton aphid that their intrinsic rate of increase and fecundity decrease significantly in the first and second generation. The aphids feeding on Bt+CpTI cotton showed higher FA and their feeding behavior detected by EPG is different significantly with Bt-cotton and conventional cotton. The possibility of FA as a sensitive indicator for the ecological risk and environmental stress of GM crops are discussed.

Genetic diversity study of native Iranian silkworm using ISSR markers

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Five diverse strains of the native Iranian silkworm were analysed using the simple sequence repeat anchored polymerase chain reaction. Samples (30 larves from 3rd day of 5th instar for each population) were collected from silkworm research institute. DNA was extracted from sericigene glands by modified Suzuki method. A set of three 5'-anchored repeat primers amplified a total of 211 bands out of which 208. A denogram constructed using the UPGMA method revealed two distinct groups. Maximum unbiased genetic distance was between Pink and Gilani and Minimum was between Gilani and Baghdadi. Gilani and Baghdadi were in one group and other populations were in separated groups. Maximum and minimum gene diversity average was in Khorasani and Baghdadi respectively. Genetic diversity average for total populations was 0.3575. Average of effective number of alleles was maximum in Khorasani and minimum in Baghdadi and for whole populations was 1.5995. Maximum polymorphism was seen in Khorasani and minimum was in Limon and Baghdadi. These results suggest that ISSR method is potentially useful for study of genetic diversity of silkworm and as a mapping tool in the silkworm.

Ecology of *Helicoverpa* spp. (Noctuidae) and their parasitoids, in relation to the use of transgenic cotton and associated refuge crops.

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Transgenic (Bt) cotton varieties provide a substantial basis for economically and environmentally sustainable insect pest management within the Australian cotton industry. Whilst the introduction of Bt cotton (in 1996) has significantly reduced pesticide use and encouraged a greater emphasis on the management of beneficial invertebrates in pest control, its impact on the overall abundance of the target moth pests, *Helicoverpa* spp., in cotton-growing regions is poorly understood. This poster will present analyses of long-term data sets from northern New South Wales and southern Queensland, including pheromone and light trapping programs, to test if populations have changed over time. In addition, the efficiency of designated refuge crops (no Bt use), which must be grown in partnership with Bt cotton to generate large numbers of *Helicoverpa* that are susceptible to Bt and thus reduce the likelihood of the development of resistance, will be discussed. Finally, the degree to which refuge crops, other agricultural crops besides cotton, and non-agricultural habitats produce significant populations of natural enemies of *Helicoverpa* will be described.

Toxicity of cascade (flufenoxuron) to natural enemies and influence upon arthropod pests in citrus orchard

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The toxicity of cascade (flufenoxuron 100 CE; 30 mL/100 L) and mixture with pyridaben 200 CE (50 mL), abamectin 18 CE (20 mL), acrinathrin 50 SC (5 mL), propargite 720 CE (50 mL) and dinocap 369 CE (50 mL) were evaluated to the predators mites *Euseius citrifolius* and *Iphiseiodes zuluagai* (Acari: Phytoseiidae), the ladybugs *Pentilia egena*, *Cycloneda sanguinea* and *Azya luteipes* (Coleoptera: Coccinellidae) and to the lacewings *Chrysoperla externa* and *Ceraeochrysa cubana* (Neuroptera: Chrysopidae) in citrus orchard. The density of preys was also evaluated between 3 and 60 days after the application, on a randomized blocks with 10 treatments, 5 replications and plots of 12 plants. The predators mites were evaluated on internal leaves and the leprosis citrus mite, *Brevipalpus phoenicis* (Acari: Tenuipalpidae) on fruits. The ladybugs and lacewings were evaluated on leaves infested with the scales *Selenaspidus articulatus* and *Parlatoria ziziphi* (Hemiptera: Diaspididae) around the tree canopy. Flufenoxuron in mixture was highly efficient to control *B. phoenicis*, as the standard mitecide fenbutatin oxide 500 SC (80 mL). The density of *S. articulatus* and *P. ziziphi* were similar to the check plants. The mixtures with pyridaben, acrinathrin and propargite were of low toxicity (<50% of reduction) to the natural enemies.

Microencapsulated α-cypermethrin and PBO with delayed release of active ingredient restores pyrethroid susceptibility in B-biotype *Bemisia tabaci*.

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Pyrethroid resistance in B-biotype *Bemisia tabaci* field populations is primarily a consequence of overproduction of esterase isoenzymes that metabolise and sequester pyrethroid insecticides. Laboratory and field studies demonstrated that the synergist piperonyl butoxide (PBO) can potentiate pyrethroid mortality against resistant field populations in Australia. This is dependant upon a delay in applying the active (α-cypermethrin), after PBO application. PBO inhibits the esterase isoenzyme, thus regaining susceptibility, by preventing pyrethroid detoxification. A single application of a novel formulation of microencapsulated α -cypermethrin allowed time for PBO to inhibit the esterase prior to the release of pyrethroid. In laboratory bioassays, using a pyrethroid resistant field population, use of microencapsulated α-cypermethrin with PBO reduced the resistance factor from 200 fold to 5 fold. A simultaneous application of PBO and α–cypermethrin only reduced the resistance factor to 145 fold. A similar gain in efficacy was also obtained in a field trial. The microencapsulated formulation gave almost complete mortality in the sprayed area, whereas α-cypermethrin applied at the registered rate achieved no mortality. The use of this microencapsulated pyrethroid and PBO clearly has the exciting potential to restore pyrethroid susceptibility in B-biotype *Bemisia tabaci.*

Cry2Ab resistance in *Helicoverpa armigera*

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Transgenic cotton that produces Cry1Ac and Cry2Ab will be used in Australia for the management of *Helicoverpa* spp. with reduced usage of synthetic chemical insecticides, to which *Helicoverpa armigera* has already developed resistance. There is some concern about the risk of resistance to the transgenic cotton by *H. armigera*, which has a demonstrated capacity for resistance to Cry1A toxins. We tested the potential for Cry2Ab resistance in *H. armigera* by selection of field populations on artificial diet containing Cry2Ab. Independently, resistance to Cry2Ab was detected in an F_2 screen involving only 33 single pairs (1/132 alleles; R. Mahon, pers comm.), suggesting that the resistance allele may be common in *H. armigera*. Shortly thereafter, resistance was detected in our strain after only three generations of selection. Following a further four generations of selection, the resistance ratio stabilised at 80-fold, a level at which Cry1Ac-resistant *H. armigera* can complete development on Cry1Ac-cotton. Preliminary characterization of this strain will be discussed.

Biochemical and molecular mechanism of prothiofos-resistance in the diamondback moth, *Plutella xylostella* (L.)

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To understand resistance mechanism of the diamondback moth (DBM, *Plutella xylostella*) against prothiofos, a resistant strain (DBM-R) has been established from the susceptible (DBM-S) strain and maintained over 100 generations under the selective pressure of prothiofos. The activity of general esterase in the DBM-R strain was 2 times higher than that in DBM-S strain, whereas there was no significant difference in glutathione S-transferase activity. Acetylcholinesterase (AChE) of the DBM-R strain, however, showed a low affinity for acetylthiocholine iodide, and its activity was 1.2-fold higher than that of the DBM-S strain. Inhibitory kinetic analysis of the resistant AChE with paraoxon exhibited the reduced bimolecular reaction constant (k_i) by 7-fold, indicating that the resistant AChE is much less sensitive to paraoxon. The nucleotide sequences of the *AChE* gene in both strains showed polymorphism due to amino acid substitutions at several positions. In addition, *AChE* gene in genomic DNA has no intron and of different length, such as 1717- and 1917-bp from DBM-S and 1614- and 1917-bp from DBM-R, showing possibly multiple copies of *AChE* genes in their genome. These results suggest that the increased AChE activity, AChE insensitivity and polymorphisms in AChE amino acid sequence contribute to the development of prothiofos-resistance in the DBM-R strain.

Analogues of piperonyl butoxide and their efficacy against metabolic enzymes.

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Analogues of piperonyl butoxide (PBO) were synthesised and structures confirmed by NMR and mass spectrometry. Homologues contained alterations to either the methylenedioxyphenol moiety or the polyethoxy side chains, and the *in vitro* efficacy of the analogues against non-specific esterases and microsomal oxidases assayed biochemically. It was confirmed from these assays that the methylenedioxyphenol ring must be intact for efficacy against microsomal oxidases, but was found to be non-essential for activity against non-specific esterases. Such analogues, therefore, are of great interest in exploiting the contribution of metabolic enzyme systems in insecticide resistance.

Insecticide defenses in *Ochlerotatus sollicitans* (Walker) (Diptera: Culicidae) larvae

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Oc. Sollicitians is a major pest mosquito along the eastern coast of North America. It is a competent vector of the eastern equine encephalitis virus (EEEV). In New Jersey, *Oc. Sollicitians* is controlled in its larval stages by temephos (an organophosphate), which has been used as primary larvicide on the salt marshes since the 1970's, and methoprene, and in the adult stage by malathion (an organophosphate) and synergized pyrethoids. Resistance of *Oc. Sollicitians* in several localities toward these insecticides has been well documented. However, the mechanisms are unclear. *Oc. Sollicitians* populations from South New Jersey and North New Jersey still have considerable sensitivity to temephos, carbaryl and resmethrin; the southern mosquitoes are less sensitive than the northern ones. Environmental factors including salinity and water pH do not significantly affect the toxicities. Toxicity assays with synergists (piperonyl butoxide and DEF) show that cytochrome P450s and esterases are the two primary enzyme systems involved in temephos, carbaryl and resmethrin detoxification. RT-PCR and molecular cloning show that many cytochrome P450s are expressed. Comparison of the cytochrome P450 profiles between the northern and southern NJ mosquito populations will be used to identify the isoform(s) responsible for the different sensitivities to the tested insecticides.

Impact of *Bacillus thuringiensis* transgenic maize on non-target arthropod populations in the southeastern United States

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Transgenic field maize has been genetically engineered with the gene from the *Bacillus thuringiensis* var. *kurstaki* (Bt), which secretes proteinaceous endotoxins that is toxic to certain species of lepidopteran pests. The effects of transgenic Bt maize containing the MON810 event on non-target arthropods were compared to those in conventional, near isogenic maize. The study was conducted at two locations in Georgia in 2001 and 2002 using visual counts, pitfall traps, and ear insect evaluations. The only insect whose numbers and mortality was consistently affected by the Bt maize was the corn earworm, *Helicoverpa zea* (Boddie), a target insect. There were no consistent effects of the Bt toxins on non-target phytophagous and predaceous arthropods observed in the visual counts, nor on those non-target arthropods recovered from the pitfall traps and the maize ear dissection. From these results, transgenic Bt field maize containing the MON810 event does not have an adverse effect on non-target phytophagous or predaceous arthropods.

Uptake and persistence of imidacloprid in California vineyards

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The glassy-winged sharpshooter (GWSS) *Homalodisca coagulata* Say has become the principal vector of the plant pathogenic bacterium *Xylella fastidiosa* Well in southern California. Strains of *X. fastidiosa* cause several economically important plant diseases, including Pierce's Disease (PD) in grapevines. The GWSS transmits this disease while it feeds on the xylem fluid of host plants. The Temecula Valley viticultural region in southern California was severely affected by a PD epidemic that began in 1996, with some wineries reporting losses of 20% to 30% of their vines. There is no cure for PD and infected vines must be removed from the vineyard. In a comparison of Admire application rates in Temecula Valley vineyards, a 16 oz. per acre treatment proved very effective at attaining xylem sap imidacloprid levels that were necessary to protect vines from sharpshooter feeding. In contrast, the same rate applied to table grapes in Coachella Valley vineyards produced levels of imidacloprid within the xylem sap that were extremely short-lived, raising concerns about the utility of this material within this region. Information about the soil type was found to be particularly important for the effective deployment of Admire applications in both regions."

Monitoring resistance to insecticides of diamondback moth and some alternate application in Thailand

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Monitoring resistance of diamondback moth, *Plutella xylostella* L. was done in the laboratory by leaf dipping bioassay. Larvae from 4 parts of Thailand; north , north- east , central and west were tested with some insecticides and *Bacillus thuringiensis* (Bt) both var. *kurstaki* and *aizawai*. Studies in 1997-2001 , larvae from central with many vegetable growing areas ; Pathumthani (PNS)and Nonthaburi province(BBS and SNS) developed resistance to almost all insecticides used by crucifer growers such as permethrin, deltamethrin, lambda-cyhalothrin, prothiofos, enofos, abamectin, fipronil, chlofenapyr and chlorfluazuron with resistant ratio(RR) 23.19-138.27 fold compared to susceptible strain from Japan . Because of the continuous use and almost same crop grown in same area throughout the year. However, in some northern area;Sunsai, Chiengmai province (SSS) had no resistance to deltamethrin, lambda-cyhalothrin, enofos, chlorfluazuron, abamectin, fipronil, chlofenapyr and spinosad with RR 3.71-10.54 fold . Every parts showed high resistance to permethrin but did not have any Bt resistant. In 2003, alternate application trial was done in farmer's field ,growing Chinese Kale, at Bangbuathong(BBS,central part). Spinosad 2 times alternated with Bt.a 2 times each sprayed day after 2 days interval throughout the crop season gave highest marketable yield and significantly lowest number of larvae.

Cloning of vitellogenin cDNAs of the oriental fruit fly, *Bactrocera dorsalis* (Hendel), and its expression analysis

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In the present research, vitellogenin (Vg) genes of the oriental fruit fly, *Bactrocera dorsalis* (Hendel) were investigated. Referring the conservation regions by alignment of several known dipteran vitellogenins (yolk proteins), a pair of degenerate primers was designed. Based on the degenerate primers, two different putative vitellogenin cDNA fragments were obtained by RT-PCR, and then the full length cDNAs corresponding to both fragments were further achieved by using 5'- and 3'-end RACE methods, cloned and sequenced. These two cDNA were named *Bdvg1* (accession No. AY368053) and *Bdvg2* (accession No. AY368054), respectively. The cDNA of *Bdvg1* was 1507 bp and contained an ORF encoding 437 amino acids, and *Bdvg2* was 1434 bp and contained an ORF encoding 440 amino acids as well. Both *Bdvg1* and *Bdvg2* were highly similar to the Vgs of relative insects such as *Ceratitis capitata*, *Anastrepha suspense*, *Calliphora erythrocephala* and *osophila melanogaster*. RT-PCR analysis revealed that both Vg genes were expressed specifically in the adult females but not in adult males, eggs, larvae and pupae. The *Bdvg1* and *Bdvg2* mRNAs were detected in the fat bodies of 1- and 2-day-old females, respectively. In addition, the Vg genes were expressed in the ovaries since day 2 after female emergence.

Diminished egg size in fenvalerate resistant strains of the diamondback moth (DBM)

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Female adults of *Plutella xylostella* (DBM) treated with sublethal doses of a pyrethloid, fenvalerate lay small-sized eggs. In the process of resistance development, DBM may also lay small-sized eggs under successive selection with insecticides at sublethal doses. In the present study, we conducted a selection experiment using DBM and fenvalerate. DBM was selected with fenvalerate at the dose of LD_{50} for 8 generations. The LD_{50} value rose gradually and resistance ratio became 60-folds. The egg size of the selected strains became significantly smaller than that of the non-selected strains within 2 or 3 generations and it stabilized thereafter. To investigate the genetic changes in the egg size in the selected strain, offspring of the selected and non-selected strains were individually reared at every other generation. The eggs of the selected strain were significantly smaller than those of the non-selected strain in all generation, and survival rate during the immature stages in the former was lower then that of the latter. Other life history traits did not differ between the two strains. This result the successive selection with fenvalerate at sublethal does yielded a fitness disadvantage in DBM population.

Aldicarb and its sulfoxide and sulfone metabolites residues in coffee leaves and efficacy for the control of the coffee leafminer

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Residues of aldicarb and its sulfoxide and sulfone oxidative metabolites in coffee leaves (*Coffea arabica* L. cv. Mundo Novo) were determined by gas chromatography, by using a NPD detector. The action of the insecticide on the control of the coffee leafminer, *Leucoptera coffeella* (Guérin-Mèneville, 1842) (Lepidoptera: Lyonetiidae), one of the main insect pest of coffee crops in Brazil, was also evaluated. The insecticide Temik 150 G was applied in December of 2001 at the dosages of 2.25 and 4.50 g of a.i./tree, in the State of São Paulo, Brazil. Twig samples were collected at zero, 30, 60, 90 120, 150, 180, 210 and 240 days after the application (DAA), in three heights of the trees: botton, middle height and top. Residues of the parent compound were not found in any leaf sample. Metabolites residues levels declined starting from 30 DAA, and were below the limit of quantitation (LOQ=0.5 mg.kg^{-1}), after 120 DAA, for aldicarb 2.25 g of a.i./tree and for its sulfoxide at 4.50 g of a.i./tree. The sulfone of this latter treatment, was found up to 150 DAA. Aldicarb at the higher dosage controlled coffee leafminer from 60-180 DAA; at the lower dosage was effective from 60-120 DAA.

Pirimiphos-methyl residues in wheat grain and in some of its processed products and the insecticide action on the control of the maize weevil.

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The degradation/persistence of residues of the organophosphorous insecticide pirimiphos-methyl was determined in wheat grain and in some of its processed products such as bran, flour, whole wheat flour and bread. The action of the insecticide on the control of the maize weevil, *Sitophilus zeamais* Mots. (Coleoptera: Curculionidae), was also evaluated. The grains were treated with the concentration of 12 $mg.kg^{-1}$ (ppm). Samples were taken at zero, 15, 30, 60, 120 and 240 days after treatment. Quantitative measurements were made by gas chromatography, using a flame photometric detector. For pirimiphos-methyl residual action studies, grain samples were taken at 15, 30 days after treatment and subsequently at monthly intervals up to one year. Treated and non-treated (control) grains were infested with weevil adults. Mortality was checked 15 days after exposure. Pirimiphos-methyl residues in grain and in wheat products opped 4 to 7 times during the sampling period. Processing operations decreased insecticide contamination in processed products: as compared to grain, bran (richer in oil) concentrated about 2.5 times more residues; whole flour had approximately the same; white flour 60% and bread 45%. The insecticide remained effective against the pest over the entire period of observation (1 year).

Action of pirimiphos-methyl on the control of the maize weevil and its residues on/in corn, popcorn grains, and in some of their processed products.

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The degradation/persistence of residues of the organophosphorous insecticide pirimiphos-methyl was determined in corn and popcorn grains and in some of their processed products such as grain, bran, hominy and corn flour from corn and prepared popcorn and grain from popcorn. The action of the insecticide on the control of the maize weevil, *Sitophilus zeamais* Mots. (Coleoptera: Curculionidae), was also evaluated. The grains were treated with the concentration of 12 mg.kg^{-1} (ppm). Samples were taken at zero, 15, 30, 60, 120 and 240 days after treatment. Quantitative measurements were made by gas chromatography, using a flame photometric detector. For pirimiphos-methyl residual action studies, samples of both types of grain were taken at 15, 30 days after treatment and subsequently at monthly intervals up to one year. Treated and non-treated (control) grains were infested with weevil adults. Mortality was checked 15 days after exposure. Pirimiphos-methyl residues were not persistent in either types of grains or their processed products. Higher residues in corn bran than in grain (1.7 time) are related to the higher oil content in that processed product. The insecticide remained effective against the pest over the entire period of observation (1 year) for both types of grains.

Effects of transgenic field corn expressing two lepidopteran-specific insecticidal proteins on the invertebrate community

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A large-scale field study during 2000-02 determined the effects of transgenic field corn expressing two Bt-derived lepidopteran-specific insecticidal proteins, VIP3A and Cry1Ab, on the nontarget invertebrate community. The experimental design compared the transgenic hybrid to a non-transgenic, isogenic hybrid with and without insecticide treatments. The abundance and diversity of foliage-dwelling and soil surface fauna were monitored by visual inspections of plants, pitfall traps, sticky cards, and litter extractions throughout the growing season and post-harvest. Aside from anticipated effects on target pest species, the transgenic hybrid had no significant negative effects on invertebrate communities. Some changes in certain taxa did occur but were linked to plant-mediated factors and the absence of feeding injury by target pest species. As expected, communities of nontarget invertebrates in the insecticide sprayed non-transgenic plots displayed significant changes in species abundance, diversity, and trophic interactions. However, communities during the third year with only non-transgenic corn were essentially the same, suggesting that disturbances associated with the insecticide may be short-lived.

Pyrethroid insecticide resistance and treated bednets efficacy in malaria control

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Metabolic and k resistance mechanisms were characterized in An. stephensi and this easily colonized species was used to evaluate the efficacy of ITNs. Bioassays performed on 4th instar larvae showed a resistance ratio of 182. The resistance was partially suppressed by PBO (54%) and DEF (51%) or both (68%). Biochemical assays revealed that esterases, monooxigenases and glutathione S-transferase levels were 1.72, 1.86 and 1.88-fold respectively in the DUB-S compared to the susceptible strain Beech. The overall permethrin metabolism rates by DUB-S mosquitoes measured by HPLC were 1.5-fold more than by Beech strain. The classic k mutation was detected in II6 of the para-type sodium channel gene from DUB-S strain. Bioassays performed on the resistant DUB-S mosquitoes using different pyrethroid-treated bednets showed that except for deltamethrin and a-cypermethrin treated bednets, the rest of the tested pyrethroid-treated bednets at standard WHO recommended doses did not produce adequate mortality rates. The deterrency test performed on susceptible and resistant An. stephensi showed that there are significant differences between the entry rates of susceptible and resistant mosquitoes into the exposure tube containing permethrin treated bednet. These data show that when mosquitoes have both k-type and metabolic resistance mechanisms, the efficacy of pyrethroid-treated bednets is questionable.

The effect of *Bt*-corn on soil invertebrates and decomposition rates of corn post-harvest residues under field and laboratory conditions.

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Effects of *Bt* corn on post-harvest residues decomposition and soil fauna, were studied in two field experiments in the Czech Republic. At each experiment location, *Bt* corn and the same cultivar of non-*Bt* corn were each planted on half of the field, and repeated at both location for three years. Field microcosms containing litter bags of *Bt* or non-*Bt* corn post harvest residues and matching field soils were exposed in matching field plots for 2 years as completely randomized blocks. Decomposition of litter bag contents and abundance of soil fauna in whole microcosms were determined. No significant effects of *Bt* corn on investigated parameters were recorded.
Either fresh post-harvest residues or post-harvest residues exposed in soil for 60 days from the above fields were used to study the effect of *Bt* corn on population growth of *Enchytraeus crypticus* (Oligochaeta: Enchytraedae) in laboratory tests. Significant (about 30%) reductions of *E. crypticus* population growth on fresh *Bt* corn litter in comparison with non *Bt* corn was observed. However, this was not observed in litter exposed in soil for 60 days. In conclusion, *Bt* corn may have a deleterious effect on decomposers in the laboratory, but these effects were minor and restricted to the initial stages of decomposition and undetectable in long-term field experiments.

Selectivity of envidor (spirodiclofen) to some natural enemies in citrus orchard.

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The trial was conducted from January to February of 2003, Taquaritinga, São Paulo, Brazil. The purpose was to evaluate the impact of spirodiclofen, in two dosages, on the beneficial arthropod fauna found in citrus orchard. The statistical design was based on completely randomized blocks, with 5 treatments and 4 replications and plots with 2 plants for the evaluations. The treatments were: spirodiclofen (Envidor, 20 and 25 mL/hectoliter), azocyclotin (Caligur, 50 mL/hectoliter), and acrinathrin + Hexythiazox (Rufast + Savey, 5 mL + 1.5 g/hectoliter). Each plot had 2 plants treated and insect density evaluated. The arthropod predator densities were evaluated at 0, 1, 4, 7, 14, 21, 29 and 40 days after the mitecides sprays. The toxicity classification was based upon Hassan's method: none ≤ 25%, low 26-50%, moderate 51-75%, and high 76-100%. The most abundant insect predator found was the Coccinellid ***Pentilia egena*** **(Coleoptera: Coccinellidae), and the predator mites found were** ***Euseius citrifolius*** **and** ***Iphiseiodes zuluagai*** **(Acari: Phytoseiidae). By the results and the trial conditions, it was concluded that spirodiclofen presented low toxicity to the predator insects, but due to the mitecide effect of the product, it was of moderate toxicity to the predator mites.**

Developmental rates and population growth of insecticide resistant and susceptible populations of the maize weevil, *Sitophilus zeamais*

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Two experiments were carried out to assess the developmental rate and population growth of four populations of *Sitophilus zeamais*. Two of these populations are resistant to pyrethroids (Jacarezinho and Juiz de Fora), while the other two are susceptible (Bragança Paulista and Sete Lagoas). The first experiment, with progeny removal and assessment every other day, allowed the estimation of the developmental rates of the four populations. The second experiment, without progeny removal and with monthly assessments, allowed the estimation of population growth and grain loss caused by each population. Both susceptible populations and the resistant population from Jacarezinho showed similar developmental rates, population growth and grain loss. The resistant population from Juiz de Fora showed reduced and delayed emergence. This population also showed smaller rate of population growth and high mortality at high densities associated with reduced grain loss. The Jacarezinho population have been maintained without insecticide selection for over a decade suggesting that resistance is fixed in this population explaining its good demographic performance, unlike what happened with the Juiz de Fora population, which was recently collected in a grain mill. Pyrethroid resistance in the Juiz de Fora population seems unstable without insecticide selection, which will likely prevent its fixation.

Partial characterization of glutathione *S*-transferases and phosphotriesterases in pyrethroid resistant and susceptible populations of the maize weevil, *Sitophilus zeamais*

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Glutathione *S*-transferases and phosphotriesterases from a susceptible population and two pyrethroid resistant populations (Jacarezinho and Juiz de Fora) were characterized through *in vitro* colorimetric assays. Glutathione *S*-transferases showed higher activity peaks at pH 9.0 and 30^{o}C, while phosphotriesterases showed higher activity peaks at pH 7.0 and 8.0 and 30^{o}C. The Km-values for glutathione *S*-transferases for the substrate CDNB were 1.3 and 1.7x higher in the susceptible population than in the resistant populations from Jacarezinho and Juiz de Fora, respectively. The Km-value for the susceptible population (49.47 ± 0.50 mM) was 7.2x higher than the Km-value for Jacarezinho (6.81 ± 0.24 mM), when the CDNB conjugation was fixed and the concentration of reduced glutathione varied. Similar trend was also obtained for the substrate DCNB. For phosphotriesterases the susceptible population showed a Km-value about 3x higher than the Km-value for both resistant populations. Therefore, the results obtained through the enzyme kinetics characterization of glutathione *S*-transferases and phosphotriesterases indicate higher catalytic efficiency of these enzymes in exercising their biological activity in the resistant populations. These findings suggest that enhanced metabolic detoxification is an important insecticide resistance mechanism against pyrethroids in *S. zeamais* and that glutathione *S*-transferases and phosphotriesterases are involved in this mechanism.

Partial life table of cotton aphid *Aphis gossypii* in transgenic Bt cotton field

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Partial life tables of cotton aphid *Aphis gossypii* were studied from mid to late August, 2003 in transgenic Bt cotton NuCTON33B field, compared to those in non-transgenic cotton Simian-3 field. The experimental fields were sprayed once with 0.36 matrine (diluted 1000 times) on 28 June to control the cotton leaf bug, and there were no other chemical spray during the growing season. Four types of nylon screen bags (10, 40, 120 and 200 meshes per square centimeter, respectively) and one uncovered control were set up to determine the magnitude of mortality factors, such as the Bt protein within plants, parasitization and predation. The results showed that (1) Compared to the uncovered control, mesh bags of different sized meshes (from 10 to 200 meshes per square centimeter) all decreased the control efficiency of natural enemies on cotton aphids. (2) Death of cotton aphids in Bt cotton (NuCTON 33B) and non-transgenic cotton (Simian-3) did not differ much, which meant that Bt cotton had little effects on cotton aphid. (3) The main factors affected the population dynamics of cotton aphid were weather conditions (mainly rains) and natural enemies (including parasitoids –Aphidiidae, Aphelinidae, and predators – ladybirds and Orius). (4) The oviposition abilities of cotton aphids on Bt cotton (NuCTON 33B) and non-transgenic cotton (Simian-3) did not differ.

A microsomal esterase confers insecticide resistance in *Spodoptera littoralis*

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Spodoptera littoralis (Lepidoptera: Noctuidae) (Boisduval), the Egyptian cotton leafworm, is a major pest of cotton and vegetable crops in Mediterranean Europe and Africa. A field strain collected from cotton at Adana in Turkey was found to have a resistance factor of 8-fold against chlorpyrifos by leaf dip bioassay. Preliminary studies of AChE sensitivity against chlorpyrifos-oxon indicated no evidence of insensitive AChE. An enhanced microsomal esterase was revealed that correlated with the observed resistance when compared to a susceptible standard strain. We report here on the purification and kinetic parameters of this microsomal esterase.

Cloning and sequencing of the latrophilin-like receptor from the economically important pest *Lucilia cuprina*.

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The latrophilin/CIRL receptor in vertebrates mediates the binding of Black Widow spider venom. We have cloned and sequenced the gene encoding a latrophilin–like receptor from *Lucilia cuprina.* Our analysis predicts a protein with 70% homology to the N-terminal and 50% to the COOH terminal region of CG8639, a hypothetical gene in the *osophila melanogaster* sequence database. CG8639 was previously identified as the *D. melanogaster* homolog of the latrophilin receptor by BLAST. The *L. cuprina* latrophilin-like receptor is expressed in all developmental stages of *L. cuprina* and is localised to the neuromuscular regions of larvae by *in situ* hybridisation. It is hypothesized this receptor plays a role in signalling / signal transduction in mammalian species, but the role in insects is largely unknown.

Insecticide resistance in *Aedes aegypti* during dengue epidemics in Taiwan, 2002

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Pest control operators were very successful in the insecticides application for mosquito control when dengue fever cases occurred in Taiwan in last twenty years. But the dengue was out of control in 2002. There were 5316 confirmed cases reported. We used 7 kinds of WHO insecticide-impregnated papers, 0.1% propoxur, 1% fenitrothion, 0.15% cyfluthrin, 0.5% etofenprox, 0.05% lambdacyhalothrin, 0.75% permethrin and 0.05% deltamethrin, to determine the insecticide susceptibility of adult *Aedes aegypti* in 2002. The resistance ratios of adult mosquitoes of the Chianjen, Lingya (2002), and Lingya (1900R) strains for permethrin compared to susceptible strains were above 312.5 folds, while those of the 4th instar larvae were 20.8, 37.5 and 102.1, respectively. Results of a synergism study on larvae resistant to permethrin showed correlations to the activities of microsomal monooxygenases and esterases.

The susceptibility of *Aedes aegypti* from northern Queensland to synthetic pyrethroids

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Aedes aegypti is the only mosquito vector of the dengue virus in northern Queensland. Despite complete reliance upon synthetic pyrethroids to control adult *Ae. aegypti* during dengue outbreaks, little is known regarding the susceptibility of *Ae. aegypti* from northern Queensland to these insecticides. Insecticide exposure bottle assays and biochemical microplate assays were conducted to examine the susceptibility of *Ae. aegypti* to synthetic pyrethroids. The susceptibility of five strains of *Ae. aegypti* from northern Queensland to five commonly used synthetic pyrethroids was compared with that of an insecticide-susceptible laboratory strain. All strains tested were fully susceptible to the synthetic pyrethroids used by mosquito control authorities in northern Queensland (lambda-cyhalothrin and deltamethrin). However, some wild strains showed significantly reduced susceptibility to permethrin, bioresmethrin and alpha-cypermethrin. Biochemical microplate assays were used to measure the activity levels of two types of enzymes (β-esterases and monoxygenases) that often confer resistance to synthetic pyrethroids. Elevated esterase activity was detected in three wild strains relative to the reference strain, but elevated oxidase activity was not detected in any of the mosquito strains. The presence of resistant traits in wild populations of *Ae. aegypti* demonstrated here supports the need to establish a resistance monitoring programme in northern Queensland to ensure the continued success of dengue outbreak control.

Design and production of a cell adhesive silk fibroin by transgenic technology

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Silk protein of *Bombyx mori* (silkworm) has used for medical materials for years and there are attempts to apply it for a regenerative medicine. One target of that is reproduction of joint cartilage. We already reported that cartilage-like tissue could be formed by chonocyte culture into silk fibroin sponge structure. This time, we designed and produced a novel silk fibroin in order to add cell adhesive activity. Silk fibroin of silkworm is consisted mainly of three components, fibroin-H, fibroin-L and P25. To improve cell adhesion activity of silk fibroin, we designed a fusion protein, that fibroin L-chain was linked with a part of human fibronectin and EGFP (L-hFN-EGFP). And we generated transgenic silkworm in order to produce L-hFN-EGFP as a part of silk protein. The transgenic silkworm secreted L-hFN-EGFP in silk gland and also made cocoon with L-hFN-EGFP. We tested cell adhesion activity on the dish coated with L-hFN-EGFP extracted from posterior silk gland. The number of adhered cells was greater on the L-hFN-EGFP coated dish than native silk coated dish and some cells were extended their pseudopod like cells on fibronectin coated dish. These results indicate that we succeeded to modify the nature of silk as we designed.

An amino acid substitution in insensitive acetylcholinesterase of *Tetranychus* mites.

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The insensitivity of the acetylcholinesterase (AChE) to organophosphate was first reported in the two spotted spider mite, *Tetranychus urticae*.and was suggested to be associated with some modification at the active site of enzyme. Recent molecular studies revealed that the insensitivity of insect AChEs was accompanied by some amino acid substitutions in its structure protein. We tried to sequence AChE cDNA in spider mites, *T. urticae* and *T. kanzawai*, and compared their sequences between a susceptible strain and an organophosphate resistant strain whose AChE was insensitive. The 687 amino acids of primary structure were deduced from the cDNA in both species. There were only three replacements of amino acid residues between AChEs of two species. In the AChE of the resistant strain of *T. urticae,* an amino acid substitution, Phe439Cis, was found. The residue is one of the acyl pocket lining components and might act an important role in the sensitibity of AChE to organophosphates. Furthermore, the amino acid 439 was changed to Trp in the resistant strain of *T. kanzawai*. The homologous amino acid substitution occurred in the insensitive AChEs of the green peach aphid and the *Culex* mosquito strongly suggests that the amino acid substitution in AChE of *Tetranychus* mites is the cause of the insensitivity.

Biochemical characterization of acetylcholinesterase in the oriental tobacco budworm, *Helicoverpa assulta*

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Acetylcholinesterase (AChE), the target site of organophosphate and carbamate insecticides, plays an important role in the transmission of nerve impulse in the insect nervous system. AChE activity of the Oriental tobacco budworm (*Helicoverpa assulta*) was higher in the head than in thorax and abdomen of the adult and gradually increased through the developmental stages. The optimum pH and temperature for the purified AChE was 8.5 and 35°C, respectively. The purified AChE was composed of two different molecular forms: an amphiphilic form (68%) and a hydrophilic form (32%). The affinity for acetylthiocholine (K_m) and the activity (V_{max}) of AChE were evaluated to be 0.05 mM and 2.1×10^{-2} unit/min, respectively. The AChE of *H. assulta* belongs to the *osophila melanogaster Ace*-orthologous AChE subfamily. Its deduced protein sequence showed the highest homology to AChE of the cotton bollworm, *Helicoverpa armigera*. Six amino acid differences were identified, which suggests that there is a polymorphism in *AChE* gene in this insect.

Building Arthropodan Mitochondrial Genomes Accessible Database – AMiGA

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"As mitochondrial DNA has shown to be an useful tool in phylogenetic studies, the number of completely sequenced mitochondrial genomes has greatly increased in the last few years. These sequences are available in some broad databases such as those of NCBI (http://www.ncbi.nih.gov) and in smaller and more functional organelle databases. During our work with the characterization of mitochondrial genomes of dipteran species we became aware of the usefulness of establishing a specific database for complete mitochondrial DNA sequences of arthropods. AMiGA is being constructed in a relational database system (MySQL) with the use of stantard query language (SQL) and populated with sequences and taxonomic information from NCBI databases. AMiGA aims to offer up-to-date sequences with useful tools for managing mitochondrial data. A web interface will provide to the public free access to nucleotide and protein sequences, including the sequence of the non-coding regions, physical maps, related reasearch institutions and scientific articles. It will be possible to access any of these data for any group of selected species. Codon usage and base frequencies will also be available. With AMiGA we hope to improve the access to arthropodan mitochondrial genomic data and to provide effective tools for researchers working in this field."

Esterase isoforms in the western flower thrips (*Frankliniella occidentalis*), and their inhibition by piperonyl butoxide

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We have studied the esterase banding pattern of several field and laboratory selected *Frankliniella occidentalis* (Pergande) stains. Some of the stains were collected on different locations at the SE of Spain and showed different levels of insecticide resistance. Others have been selected for resistance against some of the insecticides regularly used for western flower thrips control. Esterase isoforms of individual thrips have been separated by polyacrilamide gel electrophoresis, and stained for esterase activity with the model substrate a-NA. The banding patterns obtained from field and laboratory-selected insects have been compared with a standard susceptible strain. In addition, the effect of the metabolic synergist piperonyl butoxide (PBO) on esterase activity has been investigated. Gels incubated with 1 mM PBO showed a decrease in the intensity of all esterase bands. The inhibitory effect of PBO on a-NA activity was also evident from microplate reader assays of insect homogenates, and suggests the possibility of its use in the management of *F. occidentalis* resistance associated with esterase activity.

Tolerance to the *Bacillus thuringiensis* endotoxin Cry1Ac in a laboratory *Helicoverpa armigera* strain is based on immune induction, which is transmitted by a maternal effect.

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We crossed resistant insects from a laboratory strain of *Helicoverpas armigera* with susceptible insects and examined Bt-tolerance in neonates. We noticed significantly higher tolerance in offspring from resistant females crossed with susceptible males (RxS), compared to those from susceptible females crossed with resistant males (SxR). Tolerance in RxS offspring was not as high as for the resistant line, and for SxR offspring tolerance was somewhat higher than in the susceptible line. This suggests that in addition to a genetic element, Bt-tolerance in this strain is transmitted by a maternal effect. To investigate the possible mechanism of Bt-tolerance, we examined the induction of immune responses in susceptible larvae by a sub-lethal dose of Bt-toxin. One protein of 89 kDa in size was induced within two hours of exposure to the toxin. Protein concentration peaked after 6 hours and then declined to low levels by 21 hours. The protein has similarities to *osophila* proteins that act as pro-coagulants. These experiments indicate an immune induction of specific larval proteins, which may be involved in coagulation reactions in the hemolymph and gut lumen.

Resistance to Cry2Ab toxin in Australian populations of *H. armigera*

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Cotton expressing Cry1Ac toxin (Ingard®) has been grown in Australia for seven seasons. Ingard and limited quantities of two-toxin (Cry1Ac & Cry2Ab) varieties (Bollgard II®) were grown during the 2003-04 summer. Henceforth Ingard will be withawn and only Bollgard II will be available. The major Lepidopteran pest in the region is *Helicoverpa armigera*, a species with a demonstrated ability to evolve resistance to insecticides deployed against it. Resistance to Cry1Ac remains rare. This rarity was expected to enhance the prospect that Bollgard II would be resilient to the evolution of resistance. Recently a preliminary survey was conducted to detect alleles in *H. armigera* that confer resistance to Bt toxins. Only 132 alleles were examined in F_2 tests, however one was found that conferred high-level resistance (RR > 1000) to Cry2Ab. The frequency (1/132 = 0.008) was surprisingly high, as Cry2Ab has not been deployed in any transgenic crop grown in Australia. Preliminary analyses indicate the resistance is likely to be the result of a single gene, and that the resistance is largely recessive. From a resistance management perspective, the recessive nature is valuable; however modelling suggests the allele may be sufficiently common to increase in frequency through selection for homozygotes.

Heat stress breaks down the protective role of genetically engineered peas against the pea weevil (*Bruchus pisorum*)

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Recent studies have conclusively confirmed, through glasshouse and field experiments, that transgenic peas with the a-AI-1 inhibitor expressed in their seeds effectively control the pea weevil. Field trials with transgenic peas (a-AI-1 Laura line12–41) indicated that, relative to the non-transgenic plots, population growth rates of weevils were reduced by 97–98% in the transgenic plots. Also, germination tests of post-harvested field pea seeds from 1999 and 2000 show 90-99% germination for transgenic and 28-66% for non-transgenic seeds. Transgenic and non-transgenic plants were subjected to heat stress treatments under controlled conditions in a growth cabinet. The level of a-AI-1 was reduced on average by 36.3% in transgenic peas at 32/27°C compare to those grown at 27/22°C. Transgenic and non-transgenic pods of plants grown at 27/22°C and 32/27°C were inoculated with pea weevil eggs to evaluate whether the reduction in level of a-AI-1 in the transgenic pea seeds reduced the efficacy of the inhibitor. At the higher temperatures, 39% of adult pea weevil emerged, compared with 1.2% in the transgenic peas grown at the lower temperatures, indicating that heat stress reduced the protective capacity of the transgenic peas. It is possible that the transgenic peas may confer less effective protection in warmer regions. The temperatures used in the current experiments were consistent with the maximum temperature observed in the field, but the peas were exposed to a constant high temperature of 32°C for 12 h per day, whereas maximum temperatures in the field only occur for a short period around midday. If mean daily temperatures are at, or over, 27°C during flowering, growers may need to use insecticidal sprays to control pea weevils.

Insecticide resistance and *Culex pipiens* in Britain

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The importance of the *Culex pipiens* complex in the transmission of West Nile virus (WNV) has been further emphasised with the discovery of the ubiquitous hybrid 'bridge vector' forms of *Culex pipiens* in the US. In Britain and Europe the situation is different, but to what extent needs further investigation. In Britain, mosquitoes are rarely a pest and not normally subjected to control measures. Insecticide resistance is therefore expected to be absent unless the mosquitoes are immigrants, or the progeny of a mating with an immigrant, or they have been subjected to insecticide inadvertently. We report here examples of two, or possibly all three, of these scenarios. Organophosphate resistance mediated by overproduced non-specific esterases (ONSEs) has been studied extensively in *Culex pipiens* s.l. and therefore the origin and spread of many specific haplotypes is well known, most famously the A2B2 haplotype. Our discovery of this haplotype in a population of *Cx. molestus* in Scotland therefore marked these insects as being derived from immigrants. In contrast, multiple ONSE haplotypes observed in populations of *Cx. pipiens* from east England, where agricultural use of insecticide is very high, appear to be novel.

Characterisation of insensitive acetylcholinesterase in the cotton bollworm, *Helicoverpa armigera*

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Intensive use of organophosphates (OP) and carbamate insecticides over the past 50 years has lead to the development of resistance in many agriculturally important pest species, including the cotton bollworm *Helicoverpa armiegra* (Hubner). The enzyme acetylcholinesterase (AChE EC 3.1.1.7) is responsible for termination of nerve impulses at the post-synaptic membrane by breaking down the neurotransmitter, acetylcholine. AChE is the target-site for OP and carbamate insecticides which act by phosphorylating or carbamylating the active site of the enzyme to form 'transition state' intermediates that no longer attack and hydrolyse the AChE. The dephosphorylation or decarbamylation steps (k_3) either do not occur or are very slow such that the enzyme is not significantly regenerated. The subsequent accumulation of ACh leads to the continuous firing of the nervous system and ultimately, to the eventual death of the organism. AChE activity was detected using a fluorometric assay, and the sensitivity towards a range of carbamate and OP insecticides assayed.

Inhibition of *Helicoverpa armigera* esterase activity following *in vitro* exposure to piperonyl butoxide (PBO).

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Helicoverpa armigera is often considered the most economically important pest of cotton world-wide. In Australia, high levels of esterase-based resistance ensure widespread control difficulties. It has recently been discovered that piperonyl butoxide (PBO) is capable of inhibiting the resistance-associated esterases in this species. It was found that 4 hrs was the time taken for maximal inhibition of this esterase. It was also found that maximal mortality coincided with maximal esterase inhibition.

Overcoming pyrethroid resistance in *Bemisia tabaci* (Gennadius) (Hemiptera: Aleyrodidae) b-biotype with novel microencapsulated formulations of α-cypermethrin and piperonyl butoxide (PBO)

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Bemisia tabaci populations in many parts of the world are resistant against a range of insecticide classes, including pyrethroids. Piperonyl butoxide (PBO), a pyrethroid synergist, inhibits the non-specific esterases responsible for pyrethroid resistance in *B. tabaci* b-biotype. Recent studies in Australia have shown that splitting the application of the two compounds by applying PBO five hours before the pyrethroid application leads to effective *B. tabaci* b-biotype control. In order to overcome the problem of having to split the two applications, novel microencapsulated formulations containing α-cypermethrin and PBO have been developed. The efficacy of two of these novel formulations containing different ratios of the active compounds was evaluated against *B. tabaci* b-biotype on cotton plants under laboratory conditions in South Africa. Before spray application, the number of adults did not differ significantly between different treatment areas. Twenty-four hours after application, the novel treatments resulted in 92% and 78% control compared with 48% control for the α-cypermethrin and 36% control for the PBO treatments. The results indicate that the use of the microencapsulated formulations in the field has the potential to be highly effective in overcoming pyrethroid resistance and controlling the *B. tabaci* b-biotype.

Does the diamondback moth (DBM) population upsurge under insecticide application at sublethal dose?

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Physiological resurgence and/or hormoligosis frequently induced after application of insecticide at sublethal doses. Treatment of a carbamate, methomyl and a pyrethloid, fenvalerate to *Plutella xylostella* (DBM) at sublethal doses causes hormoligosis through increasing fecundity. More eggs were laid by treated female adults than those without insecticides at $LD_{12.5}$ or LD_{25}. However, the eggs laid by treated females at LD_{25} and LD_{50} were significantly smaller in size than those laid by untreated females. The reproductive effort (fecundity×egg size) did not differ between treated females and untreated ones. The treatment of a sublethal dose (LD_{50}) against the parent trended to affect the development and survival rate of offspring at immature stages. This adverse effect was apparent under harsh conditions of physical environment. The hatchability of smaller eggs laid by treated females at LD_{25}, tended to be lower than those by untreated females, and the difference was significant at humidity less than 30%. The survival rate of immature stages from the smaller eggs was significantly lower and development tended to prolong more than those from the normal sized-eggs at temperatures higher than 20°C. Therefore, it is doubtful that the outbreaks occur always through insecticide hormoligosis in particular under harsh field conditions.

Non-target effects of Bt cotton: natural enemy abundance and function

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Transgenic cottons expressing the insecticidal proteins of *Bacillus thuringiensis* (*Bt*) have been widely adopted by cotton producers throughout the USA to control lepidopteran pests. Beginning in 1999, a five year study was conducted in Arizona, USA to examine the comparative effects of *Bt* cotton on non-target organisms, with special emphasis on natural enemy abundance and the community function of the natural enemy complex. Combined analyses over all years indicate that the abundance of several natural enemy taxa, including some spiders, *Hippodamia convergens*, and *Nabis alternatus* were significantly reduced in *Bt* cotton compared with non-*Bt* cotton. In all cases, population reductions were less than 10%. However, the overall function of the natural enemy community, measured as rates of predation and parasitism on two key pests (*Pectinophora gossypiella* and *Bemisia tabaci*) of cotton in the western USA were unaffected in *Bt* cotton. Retrospective statistical power analyses indicated that sample sizes of the combined multi-year dataset were consistently sufficient to detect differences as small as 4-5% with greater than 80-90% power at a 95% level of probability. Results demonstrate that long-term and multi-factor studies are required to examine meaningful non-target effects in the field.

Preliminary construction of BAC contigs covering the chorion locus of *Bombyx mori*.

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The chorion gene locus of *Bombyx mori* has been extensively studied morphologically, genetically, biochemically and genomically until 1990 by the Kafatos lab and colleagues. The chorion is composed of more than 100 proteins, which are encoded by six closely related gene families, ErA, ErB, A, B, HcA and HcB, that map in two clusters on the chorion loeus of chromosome 2. (review: Eickbush and Izzo, 1995). Recently, our group has been engaged in the analysis of the *Bombyx* genome using modern genomic tools. And resources, in the present study, we are focusing on the construction of BAC contigs covering the chorion gene locus of chromosome 2 by the following strategy: (1) identifying chorion-specific EST markers by EST analysis of a follicle cell cDNA library, (2) screening BAC clones from a *Bombyx* BAC library by chorion-specific EST markers, and (3) constructing BAC contigs by fingerprinting and use of FPC software. Fingerprinting of 200 BAC clones screened by chorion gene products gave three main BAC contigs. By end-walking and PCR verification using BAC-end sequence, we could edit three BAC contigs into one contig spanning about 1 Mb which includes early, middle and late chorion gene families.

Activity of the new acaricide (etoxazole) on the citrus leprosis mite, *Brevipalus phoenicis* (Geijskes, 1939) (Acari: Tenuipalpidae) in field and laboratory conditions.

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Etoxazole (S-1283 11% SC) is a new molecule acaricide belonging to a new chemical group (Diphenyloxazoline). Etoxazole's modes of action is through the inhibition of the normal molting process, ovicidal egg activity and present a sterilizing effect on adult females. The experiment was conducted at Ilha Solteira/SP/Brazil. The activity of the etoxazole was evaluated on adults, larvae and eggs of *B. phoenicis* in field and Laboratory conditions. Etoxazole (1.1; 1.6; 2.7 and 5.5 g a.i. /100 L of water); hexythiazox (1.5 g a.i. /100 L of water) and flufenoxuron (3 g a.i. /100 L of water) were applied in field in a citrus orchard, variety Pera. The same acaricides and doses were used at Laboratory through the application on citrus fruits, demarcated with an arena of 2.5cm of diameter, containing adults, larvae and eggs of *B. phoenicis*. The results showed that etoxazole starting from the dose of 2.7 g i.a. /100 L of water showed: low direct activity on adults; mortality of 100% on larvae; viability of eggs inferior to 5%; adults infested on applied fruits in field until 42 days after treatment, it produced eggs, however, the emerged larvae die soon after the emergency.

Effect of new acetamiprid formulation (tablet) on the control of the whitefly *Bemisia tabaci* (Biotype B) (Hemiptera: Aleyrodidae) and *Aphis gossypii* (Hemiptera: Aphididae) in melon.

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The increasing search for safer alternatives for pests control, which are less aggressive for the environment, has brought a significant development of new insecticide formulations, with possibilities of use in agricultural pest management and contributing to a safer and more efficient way of pest control. The objective of this work was to evaluate the performance of the new formulation (tablet) of acetamiprid that is being developed for use in the transplant of vegetables to avoid the applications sequential of spray insecticides. The experiment was installed in field conditions, in Ilha Solteira/SP/Brazil, in melon. The treatments consisted: acetamiprid tablet applied on the soil in the bottom of the hole during the transplant of the seedlings (one tablet per plant); acetamiprid tablet associated to weekly applications of fenpropathrin and acetamiprid tablet associated the foliar application of acetamiprid. The evaluations were accomplished until 45 days after the transplant, by counting the number of nymphs of the whitefly and the number of aphids in 10 leaves per plot. The analysis of the results showed that the application of one tablet per transplanted plant was efficient in the control of the whitefly and aphids until 45 days after the transplant in melon.

Larvicidal activity of lignans identified in *Phryma leptostachya* var. *asiatica* Roots against three mosquito species

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The insecticidal activity of materials derived from the roots of *Phryma leptostachya* var. *asiatica* against third instar larvae of *Culex pipiens pallens, Aedes aegypti*, and *Aedes togoi* was examined. The biologically active constituents of *P. leptostachya* var. *Asiatica* roots was characterized as the leptostachydrol acetate and 8'-acetoxy-2,2',6-tetramethoxy-3,4,4',5'-dimethylenedioxyphenyl-7,7'-dioxabiclo[3.3.0]octane by spectroscopic analysis. The insecticidal activities of leptostachydrol acetate against *Culex pipiens pallens, Aedes aegypti*, and *Aedes togoi* were 100, 98.6 and 100% at 10 ppm. 8'-acetoxy-2,2',6-tetramethoxy-3,4,4',5'-dimethylenedioxyphenyl-7,7'-dioxa-biclo[3.3.0]octane showed weak or no insecticidal activity against three mosquito species at 10 ppm. The LC_{50} values of leptostachydrol acetate against *Culex pipiens pallens, Aedes aegypti* and *Aedes togoi* were 0.41, 2.1 and 2.3 pm, respectively.

Naturally occurring *Phryma leptostachya* var. *asiatica* root–derived compound merit further study as potential mosquito larval control agents or as lead compounds.

Bioefficacy of bifenthrin (talstar) 10 EC against chilli insect pests in irrigated ecosystem

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Chilli (*Capsicum annum* L.) I s an important commercial crop damaged by mite, *Polyphagotarsonemus latus* (Banks.), thrips, *Scirtothrips dorsalis* (Hood) and fruit borer, *Helicoverpa armigera* (Hub.) which occurs simultaneously. Bifenthrin (Talstar) 10% EC an insecticide cum acaricide was tested for two seasons at 40, 60 80 and 100 g a.i/ha along with slandered checks on chilli insect pests. Based on economic threshold level (ETL) five sprays were undertaken. Talstar at 80 g a.i/ha was found to be optimum and effective in reducing above pest population/damage below ETL level and recorded higher green chilli fruit yield.

Fenpropathrin (meothrin) 30 EC: an ideal insecticide in chilli (*Capsicum annuum* L.) insect pest management

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Yellow mite, *Polyphagotarsonemus latus* (Banks), thrips, *Scritothrips dorsalis* (Hood) and fruit borer, *Helicoverpa armigera* (Hubner) are the economically important pests on chilli crop. Any insecticide molecule having efficacy against above pests fits very well in the management. Fenpropathrin (Meothrin) 30 EC an insecticide cum acaricide was evaluated for two seasons during 2001-02 and 2002-03 at 50, 75, 100, 150 and 200 g.a.i./ha along with three standard check insecticides and an untreated control on chilli variety Byadagi kaddi. In another trial Meothrin was integrated in the schedule four times at different intervals and positions. Based on economic threshold level (ETL), five sprays were undertaken. Pooled data of two seasons indicated that Meothrin at 75 g.a.i./ha was found to be optimum and effective in reducing mite and thrips population below ETL upto seven days and Meothrin at 100 g.a.i./ha was found effective against fruit borer, *H. armigera.* Spray sequence of Meothrin at 75 g.a.i./ha followed by oxydemeton methyl 25 EC at 200 g.a.i./ha, Meothrin 30 EC at 100 g.a.i./ha, dicofol 18.5 EC at 475 g.a.i./ha and Dipel 8L at 1000 ml/ha was found to be ideal as it recorded significantly lower pest population and higher green chilli fruit yield.

Fenpropathrin (meothrin) 30EC: an ideal insecticide in brinjal (*Solanum melongena* L.) insect pest management

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Among the many pests damaging brinjal or egg plant, *Solanum melongena* L. leafhoppers, *Amrasca biguttula biguttula* Ishida, whitefly, *Bemisia tabaci* Gennadius, spider mites, *Tetranychus* spp. and shoot and fruit borer, *Leucinodes orbanalis* Guenee are the major ones in this locality. Insecticide having effect on all the pests is the urgent need of the farmers. Fenpropathrin (Meothrin) 30 EC an insecticide cum acaricide was evaluated for two seasons during 2001-02 and 2002-03 at 50, 75, 100, 150 and 200 g.a.i./ha along with three standard check insecticides and an untreated control on brinjal hybrid MEBH-11. In the second trial, Meothrin was integrated in the schedule five times at different intervals and positions. Based on economic threshold level (ETL), six sprays were undertaken. Results indicated that Meothrin at 100 g.a.i./ha was found to be optimum and effective in reducing all the pests. Spray sequence of oxydemeton methyl 25 EC at 200 g.a.i./ha followed by Meothrin at 75 g.a.i./ha, Dipel 8 L at 800 ml/ha, Meothrin at 100 g.a.i./ha, Dipel 8L at 1000 ml/ha and Meothrin at 100 g.a.i./ha was found to be ideal as it reduced pest population significantly and recorded higher fruit yield.

Insecticide mode of action classification: a key to insecticide resistance management

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The Insecticide Resistance Action Committee (IRAC) has developed a classification scheme for insecticides (and acaricides) that is based on mode of action (MoA). Experience has shown that effective insecticide resistance management (IRM) strategies seek to minimise the selection for resistance from any one type of insecticide. In practice, alternations, sequences or rotations of compounds from different MoA groups provide a sustainable and effective approach to IRM. This ensures that selection for resistance through the use of compounds in the same MoA group is minimised. It is recognized that insecticide resistance can also result from enhanced metabolism by enzymes within the pest, reduced penetration of the pesticide into the pest or behavioural changes of the pest. These resistance mechanisms are not linked to any site of action classification, but are specific for individual compounds or chemical groupings. Despite this, alternation of compounds from different chemical classes remains an entirely viable resistance management technique since such a practice will always minimise selection pressures. This poster describes the mode of action classification scheme and its use in IRM.

Impact of piraclostrobina on beneficial arthropod pests, predators and entomopathogens in citrus orchard.

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The present study was done to evaluate the impact of 2 applications of Comet (300 and 600 g of commercial product/100 liter), within a 60 days interval, compared to Recop (Copper Oxide) and Derosal (Carbendazim) upon arthropods and entomopathogens in citrus orchard. The populations of Citrus Rust Mite, *Phyllocoptruta oleivora,* Citrus Leaf Miner, *Phyllocnistis citrella,* Japanese Bayberry Whitefly, *Parabemisia myricae,* Rofous Scale, *Selenaspidus articulatus,* Black Parlatoria Scale, *Parlatoria ziziphus,* the predator mites of the Phytoseiidae and Stigmaeidae families and the predator insects of the Coccinellidae and Chrysopidae families were evaluated about the possible effects on their population densities after the fungicide sprayings. The results showed that: (1) Comet, can induce resurgence of the Citrus Rust Mite only sixty days after the second spraying; (2) Comet, Recop and Derosal do not have any influence on the predator insect and mite populations; (3) Comet, Recop and Derosal do not have any influence on the Leaf Miner and the Japanese Bayberry Whitefly and (4) Comet, Recop and Derosal have little influence on the population increasing of Rofous and Black Parlatoria Scales.

Use of image analysis to correlate insecticide symptomology and mode of action

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Today's competitive pesticide market demands the discovery of insecticides with novel modes of action. Recently, insecticide discovery has been transformed by the advent of imaging technologies, leading to a new way of studying insect symptomology that is inherently linked to the mode of action of insecticides. This study used a contact exposure of known insecticides to the larval stages of the Tobacco Budworm *Heliothis virescens* and symptoms were recorded by capturing images using a CCD camera and subsequent image processing. Timelapse video and quantitative measurements of locomotion were recorded over a time period of up to 24hours after exposure. A preliminary study using neurotoxic compounds with a range of mode of actions was investigated. The results showed that it was possible to differentiate between different insecticides by their symptoms, although this was not clear in every case where the end physiological effects were the same. This approach has potential to be employed to generate a library of symptomology profiles of known modes of action to which novel insecticides can be compared to aid the discovery of new chemical classes.

Field performance of thiacloprid against the tobacco flea beetle *Epithrix hirtipennis* (Melsheimer)

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The efficacy of thiacloprid for controlling the tobacco flea beetle *Epithrix hirtipennis* (Melsh.) (Coleoptera Chrysomelidae) was assessed in replicated field experiments in 2002 and 2003. In both years treatments included two doses of thiacloprid (81 and 101 ml ha^{-1} a.i.), a treated control (imidacloprid at the rate of 89 ml ha^{-1} a.i.) and an untreated control. In 2003 thiametoxam (50 g ha^{-1} a.i.) was an additional treatment. The 2002 trial crop, flue cured at a density of 27,700 plants ha^{-1}, received one application, while the 2003 trial crop, burley at a density of 47,500 plants ha^{-1}, received two applications at two weeks interval. Insecticide preparations were sprayed at normal volume at the rate of 1000 L ha^{-1}. Live beetles on ten plants in two central rows per plot were counted as response at intervals until one month after the applications. In both trials the number of live insects decreased to near zero in the first few days after all insecticide treatments and rose afterwards approaching the untreated control level in about two weeks. On average thiacloprid reduced counts in comparison with imidacloprid: by 11% and 17% in 2002 and by 60% and 78% in 2003 (lower and higher dose, respectively). Thiametoxam also reduced counts by 50% over imidacloprid.

Pyrethroids residual effect on *Anopheles* sp. mortality rates in the Brazilian Amazon.

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We had evaluated the efficacy of four pyretroids on the *Anopheles* mortality rates in order to verify their effect in indoor residual spraying in Amazon, Brazil, as a contribution to the malaria vector control program. The field work was carried in Pará State, Brazilian Amazon. Houses were treated with cypermethrin at a rate of 125mg i.a./m^2, deltametrina 25mg a.i./m^2, lambdacyalothrin 30mg a.i./m^2 and etophemprox 200mg a.i./m^2 . The entomological follow up was conducted during a five months period. The results showed that the insecticides had effect on *Anopheles* populations varying between 2 to 5 months depending on the insecticide. The mortality rates were variable according to the type of wall and insecticide. Insecticide applied in wood and non-plastered brick surfaces were more stable and lasted more than on bricks plastered ones. Labdacyalothrin showed shorter effect than the other insecticides, lasting up to 2 months. Etophemprox showed at least 3 moths effect and it was very effective in non-plastered brick surfaces. There was no statistic difference between the effect of deltametrina and cypermethrin in all surfaces tested and the duration of the effect was satisfactory up to 3 months.

Are opportunities for biological control of leafrollers in apples afforded by newer, selective insecticides?

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Insect species which, in natural systems are regulated at low densities by natural enemies are often at high densities in commercial apple orchards because insecticides decimate natural enemies and/or commercial orchards provide a rich and abundant resource for herbivores. It was also thought that the orchard habitat, due to its relatively low diversity, was less than optimal for many natural enemies. Recent experiments indicated that orchards are quite adequate habitats for the natural enemies. To determine the potential for the Obliquebanded leafroller (OBLR) biological control in commercial apple orchards, studies were conducted in 2003-2004 to assess parasitism and predation in orchards treated with conventional insecticides and with novel, more selective compounds. To eliminate the potentially lethal effects of insecticides trees were artificially infested with sentinel larvae using newly developed methods. Larvae were exposed for 48 hours and natural enemy inflicted mortality was recorded. Appropriate statistical methods were used to compare natural enemy levels and species diversity in the two insecticide regimes. Both the treatment and the geographical location of the orchard influenced mortality levels. *Actia interrupta* (Tachinidae), *Onchophanes americanus* (Braconidae) and *Exochus albifrons* (Ichneumonidae) were identified as dominant parasitoid species. Predacious insect species were also identified using different collecting methods.

Impact of genetically modified maize pollen containing a *Bacillus thuringiensis* toxin on honey bees *Apis mellifera*

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Maize pollen can be the major protein source for honey bees which are important pollinators in many ecosystems. Genetically modified maize containing a gene from *Bacillus thuringiensis* (Bt), that confers resistance to the European corn borer *Ostrinia nubialis*, may have a negative effect on non-target insects. We have investigated the maize variant Bt176 (Novartis) with the insecticidal Bt-toxin CryIAb regarding chronic long-term effects on honey bees. The test colonies received pollen treated with active toxin, whereas the control group received untreated pollen. Both experimental groups did neither differ significantly in mortality of adult bees nor in the number of open and sealed brood cells/colony during the six week trial period. This also applies for foraging activity for pollen and sugar water, brood development, brood rearing activity and the weight of newly emerged bees. In a second experiment, when an infection with the microsporidium *Nosema apis* occurred in the colonies, the bees of the Bt-group died significantly earlier and foraged less than the control group bees. Bees suffering from an microsporidian infection are more susceptible to CryIAb. Presumably both, toxin and pathogen, coacting can cause an enhanced destruction of the epithelial cells of the gut. However, the results indicate that the Bt-Toxin CryIAb is not toxic to healthy bees.

Performance of the Colorado potato beetle on plants with reduced and enhanced insect resistance

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Lectins belong to the battery of substances by which plants counteract attacks of insect herbivores. The gene encoding snowop lectin GNA (*Galanthus nivalis* agglutinin) was introduced into the genome of several crops with the aim to increase their resistance to insect pests. We examined performance of the Colorado potato beetle (CPB), *Leptinotarsa decemlineata*, on the GNA-potatoes in comparison with the parental non-transgenic cultivar and with a potato depleted of the lipoxygenase LOX H3. This enzyme is involved in the control of jasmonic acid methylation that initiates wound signalling leading to activation of the natural plant defence against insect herbivores. CPB larvae of the penultimate (3rd) and last (4th) larval instars were fed on potato leaves freshly cut from the non-flowering plants higher than 20 cm. One plant supported whole development of one group of larvae. The leaves (only the pinnate ones) were collected from the base to the top of the plant and care was taken to inflict minimal damage. The growth of CPB larvae on the GNA-potatoes was lower than on the control potatoes. The food consumption was similar, except for the first day of the last larval instar when it was reduced on the GNA-potatoes. The biomass increment in course of the experiment and the conversion efficiency of the ingested diet were significantly smaller in insects grown on the GNA potatoes. The pupae and adults developed normally but were smaller in size and their fecundity was reduced. Unexpectedly, there was no difference in any of these parameters between the insects grown on the control potatoes and on those fed the potatoes lacking LOX H3. We conclude that GNA restrains food utilization in CPB larvae but the effect is subtle and apparently insufficient for an economically relevant suppression of the CPB populations in the field. (The study was supported by grant 522/02/1507 from the Grant Agency of the Czech Republic).

Polymorphisms of acetylcholinesterase of the diamondback moth, *Plutella xylostella* in field populations in Korea

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Various organophosphorous insecticides have been used to control the diamondback moth (DBM) in Korea. Acetylcholinesterase (AChE) has been known to be involved in the insensitivity of organophosphorous and cabamate insecticides though point mutations. To analyze the AChE-based resistance among field populations of DBM in Korea, DBM larvae were collected from five local areas including 2 northern areas (Pyungchang and Yangyang) and 3 southern areas (Muju, Geochang and Jeju island). The characterization of *AChE* gene from each field population can be summarized as follows: (1) The *AChE* gene belongs to *osophila* type subfamily, (2) Size polymorphism of *AChE* gene: 175-, 447-, 593- and 638-amino acids, and (3) various mutations at different positions contribute to heterogeneity among field populations. These results suggest that the DBM can attain the rapid resistance to organophosphorous insecticides based on polymorphisms of *AChE* gene among field populations having different history of agrochemical treatments and various environmental factors.

Search for P450 allele associated with permethrin resistance in *Spodoptera exigua*

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Cytochrome P450 enzymes (P450) play important roles in the metabolisms of both endogenous and xenobiotic compounds. P450 have been extensively studied in insects because of their importance in the insecticide resistance mechanism and the metabolism of secondary plant metabolites. The beet armyworm, *Spodoptera exigua,* were selected in laboratory with permethrin. The selected population showed 84-fold permethrin resistance. Resistance to permethrin was decreased until 12- fold when the resistant strain (TR) was pre-treated with PBO as P450 inhibitor. This suggests that P450 is related to permethrin resistance of TR. We screened fifteen P450 genes from *S. exigua* c-DNA library. Especially, CYP9A9 gene expressed 3-fold higher in the midgut and the fat body in TR than those in the susceptible strain (TS). Western blot analysis revealed that CYP9A9 gene in midgut of TR was translated 16-fold more than that of TS. However, no difference of deduced amino acid sequence of CYP9A9 was observed between TS and TR, and the possibility of gene amplification in TR was denied by sourthern blot. It is suggested the difference of regulatory gene element involved in CYP9A9 expression between TS and TR.

Role of loop C and the loop B-C interval of the nicotinic receptor a subunit in its selective interactions with imidacloprid in insects

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Neonicotinoids including imidacloprid are agonists of insect nicotinic acetylcholine receptors (nAChRs). We have found that the recombinant *osophila* SAD-chicken b2 hybrid nAChR was more sensitive to imidacloprid than the chicken a4b2 nAChR, suggesting that region(s) interacting with imidacloprid are present in the a SAD subunit. The YXCC motif in loop C of the a subunits has been shown to form part of the agonist-binding sites. The X residue in this motif of SAD subunit is proline, whereas the equivalent residue of the a4 subunits is glutamate. Furthermore, insect a subunits have longer amino acid sequences between loops B and C than is the case for the corresponding region in vertebrate nAChRs. Thus, we have examined the roles of loop C and loop B-C interval region in the interactions with imidacloprid. The P242E mutation in loop C of the SAD subunit reduced imidacloprid sensitivity of the SADb2 hybrid nAChR, whereas the E219P mutation of the a4 subunit increased the imidacloprid sensitivity of the a4b2 nAChR. Deletion of the loop B-C interval region from the SAD subunit slightly enhanced the effect of the P242E mutation on the SADb2 hybrid nAChR, suggesting important roles of the regions investigated in the selective nAChR-imidacloprid interactions.

Tolerance of obliquebanded leafrollers to selected insecticides

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Populations of obliquebanded leafrollers, *Choristoneura rosaceana*, were sampled from orchards in British Columbia and Washington State and assayed for tolerances to insecticides. Insecticides tested included materials with a long history of use such as azinphos-methyl, and newly-introduced materials such as spinosad, indoxacarb, and the benzoylhydrazine insect growth regulators tebufenozide and methoxyfenozide. Significant differences in response to insecticides were observed across populations. Patterns of cross-resistance were determined between azinphos-methyl and other materials. Resistance management strategies were suggested for tree fruits based on these findings.

Performance of families of *Ostrinia nubilalis* (Hubner) (Lepidoptera: Crambidae) on lyophilized corn tissue: tool for selection experiments to create resistant colonies and F2 screens

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A method to use lyophilized corn tissue containing the *Bacillus thuringiensis* protein Cry1Ab is described for *Ostrinia nubilalis*. Tests were conducted on both laboratory and field-collected individuals to evaluate the bioassay for its potential use in selection and resistance detection procedures. Freeze-ying of transgenic corn leaves did not decrease the amount of Cry1Ab present when compared to frozen leaf tissue collected from the same experimental rows. In addition, incorporating the tissue into an *O. nubilalis* meridic diet was effective in killing larvae. Analyses of F_2 families originating from field-collected females indicated significant differences among families in the survival of F_2 larvae on a diet composed of 5% of its nutritive material as Cry1Ab tissue. After backcrossing F_2 individuals to a susceptible, laboratory colony we observed that the survival of BCF_2 families exposed to Cry1Ab tissue was dependent on the survival of the F_2 from which they originated. The BCF_2 family analysis indicates that the variation among F_2 families for survival on Cry1Ab has a genetic component. The bioassay using lyophilized tissue containing Cry1Ab therefore has great potential as a research tool for investigations of the genetic basis of resistance to Cry proteins and methods used to detect resistance to Cry1Ab.

Screening sugarcane clones for resistance to cane weevil borer in Viti Levu, Fiji.

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An intensive research was conducted in Lautoka during the 2000- 2003 period to develop an appropriate method of screening sugarcane clones for cane weevil borer (Rhabdoscelus obscurus, Boisd.) resistance. Results indicated that the weevil preferred to oviposit and feed more on Mana variety more than any of the other commercial varieties screened. Using the feeding preference of the weevil larvae, a larval tray screening method based on the mean length of tunnel bored per sugarcane variety was developed. The duration of the larval tray method was 30 days using cane stalks, tin trays and borer larvae. The larval tray method showed significant correlation with field results at 5% level of significance in two different locations of western Viti Levu.

Acaritouch® EC, a novel miticide which contains a food additive as an active ingredient

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We have been developing pesticides focusing attention on compounds used for food and food additives because these compounds are considered safe to the environment and human beings. Our first development is KALIGREEN® SP in which the active ingredient is potassium bicarbonate. KALIGREEN® SP has been used for the control of powdery mildew in many countries. In our recent study, we have developed a new miticide ACARITOUCH® EC. The active ingredient of ACARITOUCH® EC is a propylene glycol fatty acid ester that is used commonly as a raising agent in the baking industry. We evaluated the efficacy of more than 200 different foods and food additives against adult female two-spotted spider mites, and finally found that propylene glycol fatty acid esters showed significant efficacy. The monolauric acid ester indicates the highest miticidal activity, which shows 100% miticidal effect in concentration of over 0.25 gram per litter. Since the activity against mite's eggs is weaker than the activity against the other life stages, two times applications at intervals of 5 to 7 days can enhance the effect of ACARITOUCH® EC strongly. We sell ACARITOUCH® EC for two-spotted spider mites and citrus red mites in Japan at present.

Enhanced efficacy of various insecticides against lightbrown apple moth larvae by a horticultural spray oil.

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Lightbrown apple moth (LBAM) (*Epiphyas postvittana* (Walker)) (Lepidoptera:Tortricidae) is a serious pest of fruit crops and ornamentals in Australia and New Zealand. In citrus, the larvae feed under the calyx of young fruit, resulting in scarring (halo damage), which leads to a downgrading in price. The larvae can also infest mature fruit, causing costly delays and fumigation when intercepted on consignments sent to various export markets. Horticultural sprays oils alone and in combination with insecticides have been used to control pests in citrus IPM for many years. However, organophosphate insecticides remain a critical component to the management of LBAM in export focussed citrus production. It is important to encourage the bio-rational use of these pesticides and explore the efficacy of insecticides with novel modes of action, and, potentially, less deleterious effects on the environment and beneficial insects. In this laboratory study, a high paraffin content horticultural spray oil, Brella™, significantly increased the efficacy of traditional insecticides, methomyl and chloropyrifos. Emamectin benzoate, spinosad and indoxacarb were the most efficacious of the 'novel' insecticides tested against LBAM larvae. These insecticides also showed significant enhancement when combined with Brella. The potential implications for the field control of LBAM larvae are discussed.

Resistance-associated point mutations in acetylcholinesterase gene in the cotton aphid

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The sequences of two acetylcholinesterase (AChE) genes, *Ace1* (orthologous to *Ace* of *osophila melanogaster*) and *Ace2* (paralogous to *Ace*), from the cotton aphid (*Aphis gossypii*) were identified and compared between carbamate resistant and susceptible strains. We found no amino acid substitution in *Ace1*. On the other hand, two amino acid substitutions were detected in *Ace2*. The one substitution was found in two pirimicarb-resistant strains. This position corresponds to one of the 14 aromatic residues lining the active site gorge and located at the acyl pocket. We expected that this substitution plays an important role in pirimicarb resistance. The other amino acid substitution was located at one of the three residues which form the oxyanion hole in the active site of AChE. It remains unclear whether this substitution contributes to insecticide resistance in *A. gossypii*, although it might act a role in the sensitivity of AChE.

Action of deltamethrin on the control of the fruit small borer and its residues on/in fruits and leaves from two cultivation systems of staked tomato crops.

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The degradation/persistence of residues of the pyrethroid insecticide deltamethrin from two cultivation systems of staked tomato crops: experimental cultivation (single line) and commercial one (double line) was evaluated on/in fruits and leaves. The action of the insecticide on the control of the fruit small borer, *Neoleucinodes elegantalis* (Guenée, 1854) (Lepidoptera: Crambidae) was also determined. The treatments, with three replications each, were: A - nine (9) applications at the dosage of 1 g a.i.100 L^{-1} of water, single line cultivation system; B - the same applied as treatment A, double line; C - control. Fruit samples were taken at (-1), zero, 3, 5 and 10 days after last application; leaf samples at (-1), zero, 1, 3, 10 and 20 days. Quantitative measurements were made by gas chromatography, using an electron capture detector. For deltamethrin residual action studies 6 evaluations of the insect pest infestations were made based on percentage of fruit damage. Deltamethrin residues were below the corresponding MRL (0.03 $mg.kg^{-1}$) in fruits from commercial cultivation system (double line) at the end of safety interval (3 days). Also, the residues were higher on/in leaves as compared to fruits. The experimental cultivation system (single line) showed, in general, higher residue levels than the commercial one, giving, in consequence, a better insect pest control.

Carbaryl residues on/in fruits and leaves from two cultivation systems of staked tomato crops and the insecticide action on the control of the fruit small borer.

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The degradation/persistence of residues of the carbamate insecticide carbaryl from two cultivation systems of staked tomato crops: experimental cultivation (single line) and commercial one (double line) was evaluated on/in fruits and leaves. The action of the insecticide on the control of the fruit small borer, *Neoleucinodes elegantalis* (Guenée, 1854) (Lepidoptera: Crambidae) was also determined. The treatments, with three replications each, were: A - nine (9) applications at the dosage of 108 g a.i.100 L^{-1} of water, single line cultivation system; B - the same applied as treatment A, double line; C - control. Fruit samples were taken at (-1), zero, 3, 5 and 10 days after last application; leaf samples at (-1), zero, 1, 3, 10 and 20 days. Quantitative measurements were made by gas chromatography, using a mass selective detector. For carbaryl residual action studies 6 evaluations of the insect pest infestations were made based on percentage of fruit damage. Carbaryl residues were above the corresponding MRL (0.1 $mg.kg^{-1}$) in fruits from both cultivation systems at the end of safety interval (3 days). Also, residues were higher on/in leaves as compared to fruits. The experimental cultivation system (single line) always showed higher residue levels than the commercial one (double line), giving, in consequence, a better insect pest control.

The influence of a biotech cotton crop on arthropod biodiversity in a commercial-farm set-up in South Africa

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Cotton production in South Africa has declined over the last five years, resulting in a national crop yield that is considerably lower than present national consumption. The National Department of Agriculture, in collaboration with Cotton SA and other role-players, is therefore currently formulating a strategic plan to stabilize and expand cotton production. The introduction of cotton as a biotech crop, which enables the commercial farmer to achieve higher yields with lower input costs, will play a large role in achieving these goals. The acceptance of cotton varieties displaying both bollworm-resistant and glyphosate-resistant characteristics in a single cotton plant (so-called stack-gene varieties) by government, industry, environmental organizations and the cotton producer will depend, amongst other factors, on the influence they will have on biodiversity. Arthropod biodiversity was therefore compared within fields treated with Roundup Ready™ and Bollgard™, conventional or non-transgenic cotton, and a stack-gene variety. Preliminary results are given to provide information about the possible impact of these genetically altered products on biodiversity, for the scientific community, industry and other interested parties.

Chitinases induce unexpected effects on *Myzus persicae* Sulzer (Homoptera: Aphididae)

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As it is largely present in invertebrates but absent from plants and vertebrates, chitin appears to be a selective target for pest control agents. Chitinases (EC 3.2.1.14) hydrolyse chitin in the exoskeleton during insect moulting and in the peritrophic matrix. Such properties could be used in the development of new strategies against pests. As part of a program designed to develop potato plant resistance, an insect chitinase gene from the chrysomelid *Phaedon cochleariae* was inserted in potato plants (*Solanum tuberosum* cv. Désirée) via *Agrobacterium* gene transfer. Unexpected effects (i.e. reduction of pre-reproductive period and enhancement of daily fecundity) were observed on the peach-potato aphid *Myzus persicae* when reared on the transformed potato lines. In order to evaluate direct chitinase involvement in the physiological effects observed upon aphid feeding, a bacterial (*Serratia marcescens*) chitinase was provided via artificial diets. Ingestion of bacterial chitinase by aphids reduced nymphal mortality and enhanced the intrinsic rate of natural increase of the populations. For the first time, our results document unexpected effects of chitinase which improved several biological parameters related to *M. persicae* population's growth, a major pest deprived of peritrophic matrix. They also question the relevance of a chitinase-based strategy in a potato culture protection program.

RaPID Assay® technique to test the toxicity of spinosad in the noctuid endoparasitoid *Hyposoter didymator* (Hymenoptera: Ichneumonidae).

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Hyposoter didymator (Thunberg) is an endoparasitoid of *Spodoptera, Heliothis* and *Helicoverpa* larvae in several European countries. The use of pesticides in IPM programmes stressing biological control implies the previous step of determining the impact on the enemies. Spinosad, a neurotoxic compound obtained from a soil actinomycete, is considered a good candidate for the control of lepidopteran pests. A quantitative enzyme immunoassay using magnetic beats (RaPID Assay®) was used to determine the amount accumulated in the body of different stages of *H. didymator* after ingestion or topical exposure at concentrations of 1000 mg a.i./l. Cocoons retained the insecticide in high amounts preventing the pupae from being contaminated. Larvae and adults accumulated very low amounts, but were lethal to them irrespective of the mode of exposure. More than a half of the insecticide that remained in the adult body was found in the ovaries. The endoparasitoid seemed to be more susceptible to the insecticide than the pest.

Insecticidal activity of the lectin from ground ivy, Gleheda

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The insecticidal activity of the lectin Gleheda (related to the classical legume lectins) from leaves of the ground ivy (*Glechoma hederacea*, family Lamiaceae) has been tested against the pests *Spodoptera littoralis* and *Leptinotarsa decemlineata*. In oral treatment bioassay with larvae, Gleheda affected larval growth and halted larval feeding clearly in both species. Action of Gleheda may rely on its specific carbohydrate-binding activity, so we analyzed its activity on the insect midgut (mainly consisting of a matrix of chitin and proteins), and checked the membrane for aberrations using the scanning electron microscopy. Gleheda activity was compared to that of the GlcNAc-specific wheat germ agglutinin lectin (WGA) and the chitin synthesis inhibitor diflubenzuron. Interaction of the lectin with proteins from different insect tissues (foregut, midgut, hindgut, hemolymph, Malpighian tubules and fat body) was also evaluated. The possible mode(s) of insecticidal action is discussed.

Identifying mechanisms of cyromazine resistance in *Drosophila melanogaster*

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The insecticide cyromazine is classified as an insect growth regulator. It has been used to control pest species such as the housefly (*Musca domestica*), the Australian sheep blowfly (*Lucilia cuprina*) and the pea leaf miner (*Liriomyza huidobrensis*). The physiological effects caused by cyromazine include necrotic lesions and irregular hardening of the cuticle, elongation of the larval insect body and death. The molecular mode of action of cyromazine has not been determined. Although cyromazine resistance exists in the field, mechanisms of resistance are unknown. Chemical mutagenesis studies have identified at least four genes conferring cyromazine resistance in *Drosophila* melanogaster. One of these genes, Rst(2b)Cyr, had previously been mapped to chromosome 2R. Using polymorphic molecular markers (single nucleotide polymorphisms (SNPs), restriction fragment length polymorphisms (RFLPs) and microsatellites) and recombination mapping, we have refined the location of Rst(2b)Cyr to a region of 1 map unit which contains 74 putative open reading frames. In addition to mapping approaches, affymetrix microarrays and deficiency kit screening have been used to identify potential candidate genes involved in resistance and mode of action(s) of cyromazine.

Effects of cypermethrin and chlorpyrifos residues in cattle dung on the development of the dung beetle *Copris tripartitus* (Coleoptera: Scarabaeidae)

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Fresh dung was collected from untreated cattle and cattle dosed with a spray-on formulation of cypermethrin and chlorpyrifos (2.1g /cow) on days 1, 3, 5 and 7 post-treatment. The dung was bioassayed using the dung beetle *Copris tripartitus* Waterhouse. Residues of cypermethrin and chlorpyrifos were sufficient to inhibit oviposition by *C. tripartitus* in day 1 dung but there was no significant effect on egg laying in dung collected at days 3-7 post-treatment in sexually matured female. But, in the second generation residues of cypermethrin and chlorpyrifos were sufficient to inhibit oviposition by *C. tripartitus* in treatment after 1 and 3 days. Although there was no significantly different effect on egg laying in dung collected at days 3-7 post-treatment in first breeding but, in second breeding, toxic effect reveal in 3 and 5 days dung treatments. Nulliparous dung beetles fed for 200days on dung collected 0, 1 and 3 days after treatment of cow with cypermethrin and chlorpyrifos show little ovarian development.

Biogeography of gall-flies (Diptera: Fergusoninidae) and associated parasitoids

GS Taylor, KA Davies, J La Salle, AD Austin

Paper not available at time of production

Systematic and biological studies of larvae of the Korean species of the genus Pyrrhalta (Coleoptera: Chrysomelidae: Galerucinae)

JE Lee, JY Park

Paper not available at time of production

A systematic study on immature stages of Tipulinae (Diptera: Tipulidae) from Korea

DS Kim, JE Lee

Paper not available at time of production

Bt tolerance by an elevated immune response in Ephestia kuehniella

MM Rahman, HLS Roberts, M Sarjan, S Asgari, O Schmidt

Paper not available at time of production

Controlling white grubs Phyllophaga vetula Horn (Melolonthidae) with an entomopathogenic fungus and nematodes

J Ruiz-Vega, T Aquino Bolanos, R Perez Pacheco, C Deloya

Paper not available at time of production

Electron microscopic observation on the multiplication of chikungunya virus in Aedes albopictus

H Yamanishi

Paper not available at time of production

Parasitoids on Acronicta rumicis in South Korea

S Nam

Paper not available at time of production

Six-legged science: lessons learnt in science communication

CJ Stephens, NB Stevens

Paper not available at time of production

Efficiency Of Parasitoids From A Behavioural Perspective

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Whilst parasitoids benefit from host herbivore-induced plant volatiles, their behavioural response to these olfactory cues is variable. This variability can cause the inconsistent performance of parasitoids as natural antagonists, but it may also facilitate improvement of their efficiency in biological control through genetic manipulation. Using a model system consisting of cabbage plant *Brassica oleracea*, herbivorous insect *Pieris brassicae*, and parasitic wasp *Cotesia glomerata*, we address how genotypic differences in olfactory response influence host parasitism, and how genetic manipulation of the odour-guided behaviour can enhance the efficiency of parasitoids. Bidirectional selection resulted in two genetically diverged strains of *C. glomerata* in behavioural response to the host-induced plant volatile. Simulation experiments demonstrated that the capacity of both habitat colonization and parasitism significantly increased in the high olfactory response strain, as compared to the low olfactory response strain. Thus, the efficiency of parasitoids can be improved by genetic manipulation of relevant traits. A newly developed technique to mark parasitoids with a stable isotope makes it feasible to test this postulate in the field. The novelty of this technique allows the movement of parasitoids and the resultant parasitism to be tracked at a spatial scale.

Biological control of thrips in greenhouses: the European and Australian experience

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Western flower thrips (WFT), *Frankliniella occidentalis* is the main thrips pest of greenhouse crops in northern Europe, although other species also occur. WFT was first recorded in the UK in 1986, in other northern European countries around the same time, but in Australia not until 1993. Successful biological control strategies have since been developed for use in Integrated Pest Management programmes on many greenhouse crops. The most widely used biological control agent used against thrips in northern Europe is the predatory mite, *Neoseiulus cucumeris*, supplemented on some crops by other mite species, predatory bugs or entomopathogenic fungi. Some crops such as sweet pepper, cyclamen and other pot/bedding plants can now be grown without the use of pesticides against thrips. UK growers using biological control do not experience severe problems with the tospoviruses *Tomato spotted wilt virus* and *Impatiens necrotic spot virus*. A recent development has been the successful commercial use of entomopathogenic nematodes, *Steinernema feltiae* against WFT on chrysanthemums and other ornamentals. Australia has far fewer biological control options, but is making progress in reducing reliance on chemicals through a similar multi-pronged approach to thrips management. Current research to improve biological control methods for thrips on various crops will be discussed.

Variability in responses of discrete laboratory populations of western flower thrips, *Frankliniella occidentalis* (Pergande) to entomopathogenic fungi

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A collaborative research project was undertaken to evaluate entomopathogenic fungi against discrete populations of western flower thrips, a major pest of greenhouse and field vegetable and ornamental crops. Using a standardised laboratory bioassay technique involving a single spore deposit as a discriminating dose, entomology research laboratories in Australia, Canada and the USA are testing a range of promising fungal isolates of *Beauveria bassiana, Metarhizium anisopliae* and *Verticillium lecanii* against second instar and adult female western flower thrips. Data illustrate variability in response by different geographic strains of the target organism. Results of the research to date and future directions in the development of fungi as effective microbial control agents are presented.

Vain Hopes and Last Resorts in Field and Forest Pest Suppression

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It is tempting to look back at the techniques that have been implemented for the suppression of pests of field crops and forest systems and wonder what practitioners were thinking about. Ancient approaches relied on superstitions and divine intervention. Approaches that rely on trapping, herding, burning, barriers, or nasty combinations of highly toxic materials have been used, rejected, and resurrected. Some of these ideas worked when there were no other alternatives. Others never worked at all, but memories are short and wishful thinking or desperation can be powerful forces when the sales pitch is promising. More recently, in part because of dissatisfaction with mixed results or negative impacts of high technology, there has been a return to the spiritual approach to pest suppression and we are left wondering

Good Ideas Gone Bad in Structural Pest Control

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In recent years there has been an increasing awareness of consumers of the potential negative impact of insecticides that has resulted in an interest in the use of alternative non-toxic control strategies in urban environments. In addition to the traditional sources of pest control chemicals and equipment, interest in alternative strategies has spurned a cottage industry that markets and promotes various devices, naturally occurring plant products, and remedies designed to provide safe and effective pest control. Even though many of these strategies may be based on some scientific observations, most have not undergone rigorous scientific testing. Most of these can be conveniently organized into three distinct categories: good ideas that have gone bad, bad ideas that are really bad, and bad ideas on the verge of fraud.

Mistakes and Hyperbole in Medical and Veterinary Entomology

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Many imaginative approaches and devices have been designed and marketed for control of arthropod pests of people and animals. A sampling of these will be presented, dealing with mosquitoes, head lice, fleas, and flies. A notable mistake from the historical published literature dealing with delusory parasitosis also will be addressed.

Controlling the Diamondback moth (*Plutella xylostella*) with spinosad applied to white cabbage and cauliflower seeds.

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Three field experiments were carried out with white cabbage (2000 and 2001) and cauliflower (2000) to assess the protection against Diamondback moth with various doses of the spinosad. The plants were raised in trays of modules filled loosely with peat-based potting compost. Batches of seed filmcoated with spinosad at doses of 12, 24, 48, 72, and 96 g a.i. per 100,000 seeds were compared with untreated seeds. Filmcoating with spinosad in the doses used, resulted in the same percentage of usable plants of both crops as the untreated seeds, about 98 % at the moment of transplanting. Seeds filmcoated with spinosad appeared to reduce the number of white cabbage plants attacked by Diamondback moth caterpillars and the number of caterpillars present on the plants for at least 11 weeks after sowing. Treatment with 48 g a.i. spinosad per 100,000 seeds decreased by about 90 % the number of cauliflower plants infested with caterpillars at least 5 weeks after transplanting, inwhich is 10 weeks after sowing. The treatment with 96 g a.i. resulted in 100 % protection against the Diamondback moth.

Integrated Sugar Beet Management Research Program and practices in Iran

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Up to 200000 hectares is under sugar beet culturing in 17 provinces of Iran. The average production, 35.05 tons/hectares, shows that this crop is affected by different factors. Pests' damage is one of the most important factors. A survey of this situation was conducted to evaluate the sugar beet production information, goals and problems; sources of sugar beet IPM and use of IPM practices for producing maximum yield while keeping production and protection costs as low as possible with emphasis on ICM programs. According to this survey, 15 harmful insects, 20 pathogens and 10 kinds of weeds recorded. Some of them are key pests which decrease the total production below 50%. We have been mentioned to the IPM research program as our national purpose since 1999. Following this program the IPM practices conducted in different pilots with emphasis on good monitoring of insects, weeds and diseases; use of suitable agronomic practices; improving the chemical control methods. The results show a high decreasing in poisons application in this area.

The IPM Practices for controlling the main Sugar Beet Weevils in Iran

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Fifteen weevils' species attack to sugar beet in warm and dry regions of Iran. Two species, *Bothynoderes obliquifasciatus* Germ, *Temnorhinus brevirostris* Gyll are key pests. Their adults feed from seedlings after immigration and female laid eggs individually on the upper surface of leaves which look like a block dot. The emerged larvae make a hole in leaves then fall down to cracks in soil near the roots. Larval feeding from roots occur serious damage not only by direct infestation but also by providing suitable condition for root rots. Farmers generally spray high dosage poisons every 2-3 days in early season. In this study the IPM practices conducted base upon mechanical, chemical and agronomical control methods. It included; collecting the hibernator adults from cracks near desert weeds' roots and digging a narrow canal around fields for capturing the immigrated adults in early spring; spraying the seedlings with safe poisons against the adults and eggs; Agricultural practices like early planting , suitable weeding and good irrigation program during summer. The results show decreasing in damage.

Control of mites and insects in flower bulbs during storage: short treatments with controlled atmosphere.

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Controlled Atmosphere (CA) treatments are an attractive method for insect and mite control in flower bulbs during storage. To minimise the risk of bulb damage, the aim of this research is focused on development of short CA treatments (24 to 48 hours) to achieve complete pest control. Specific atmospheres, low oxygen or high carbon dioxide, are needed to kill different species of mites and insects. The treatment temperature is important as well as the duration of the treatment. Higher mortality rates were found when temperatures increased (25° to 30°C). Two short CA treatments with one week interval gave better results compared with the same period in one treatment. In small scale experiments the dry bulb mite *Aceria tulipae* in tulip bulbs and the bulb scale mite *Steneotarsonemus laticeps* in Hippeastrum bulbs were killed at low O_2 concentrations (Ultra Low Oxygen, <1 % O_2). The bulb mite *Rhizoglyphus robini* in lily bulbs and gladiolus thrips *Thrips simplex* in gladiolus corms were not controlled with this method. Bulb mites and gladiolus thrips were sensitive for high carbon dioxide concentrations around 30 - 60% CO_2. Advantages of this approach could be a convenient application for bulb growers because it leaves no residues on bulbs and corms. Further research is needed to determine the effects of the CA treatments on the flower bulbs and the pests in large bulb storage rooms.

Multi-species dispensers for mating disruption of codling (*Cydia pomonella* L.) and oriental fruit moth (*Grapholita molesta* Busck) on pome fruit.

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Oriental fruit moth (OFM) and codling moth (CM) are key pests of fruit grown in Victoria, Australia and in Michigan, USA. Although CM was the major pest of pome fruit, and OFM was the primary pest of stone fruit, OFM is now established as a pest of pome fruit in Victoria. In the USA and Canada OFM also damages apples, while in Chile and Argentina CM has expanded its host range to peaches. These pests have the ability to migrate and invade new host-plants and growers now need to cater for both OFM and CM together in their pest management strategies. New hand-applied dispensers for mating disruption (MD) of OFM and CM together were evaluated in counter seasonal replicated trials in Northern Victoria (Australia) and Michigan (USA). Trials were conducted for three years on randomly selected pear blocks in orchards in the Goulburn Valley, Victoria and in apple blocks in Michigan orchards where OFM and CM damaged together. Although there was a lot of variation in initial OFM and CM populations the results demonstrated that new multi-species MD dispensers performed similarly with hand applied dispensers for OFM and CM applied individually during three seasons. There were no damaged fruit found on the MD treated plots, but in the control plots without MD, fruit damage averaged at 0.2%. In general, the numbers of OFM and CM caught under all treatments were decreased at the end of the season.

First Australian trials of sprayable microencapsulated pheromone for mating disruption of oriental fruit moth (*Grapholita molesta* Busck) on peaches and pears.

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Sprayable microencapsulated pheromone products for mating disruption have an advantage in easy application with standard spray equipment and compatibility with most insecticides and fungicides. Field trials of a sprayable pheromone formulation for oriental fruit moth (OFM) mating disruption (MD) were established in 2002 for the first time in Australia and continued in 2003-04. Trials were conducted on randomly selected blocks of peaches and pears in orchards in the Goulburn Valley, Victoria. The sprayable pheromone was applied monthly for OFM control with conventional orchard spray equipment. Fruit blocks treated for MD by hand applied dispensers in the registered rate of 500 dispensers per ha were used as a standard. Fruit blocks without MD treatment, but under a conventional insecticide spray program for OFM were used as control. Preliminary results demonstrated that monthly applications of sprayable formulation on pears were as effective for OFM control as hand applied dispensers (500 dispensers per ha) if the initial level of OFM infestation was medium or low. Monthly sprayable applications on peaches were not effective enough to control medium level OFM populations. Experiments on peaches in 2003-04 with fortnightly spray intervals demonstrated no significant differences with standard hand applied dispensers. Sprayable pheromone products for MD of OFM have good potential in pheromone based IPM in Australian orchards.

Nutrient depletion in Western flower thrips affects their response to odour and visual cues

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The physiological state of insects, especially depletion of nutrients through starvation, is known to affect their behaviour. The effects of starvation on the walking or flying response of female Western flower thrips' to visual and/or odour cues in two types of olfactometer were examined. Thrips starved for 0, 1, 4, or 24 h were limited to walking in response to an odour cue (1 ml of 10% p-anisaldehyde) in a Y-tube olfactometer, while thrips starved for 0, 1, 4, 24, 48 or 72 h were able to fly in response to a visual cue (98 cm^2 yellow sticky trap) and/or odour cue (0.5 ml p-anisaldehyde) in a wind tunnel olfactometer. More thrips walked up the odour-laden arm in the Y-tube when starved for at least 4 h (76%) then those not starved (58.7 %) or only starved for 1 h (62.7%). In the wind tunnel experiments the percentage of thrips to fly (regardless of cue) or land on a yellow sticky trap changed significantly with starvation period ($P<0.001$). The percentage to fly or land on the sticky trap increased between those not starved (8% to fly, 7% on trap) and those starved for 4 h (83% to fly, 56% on trap) and decreased slightly between 48 (75% to fly, 39% on trap) and 72 h (57% to fly, 30% on trap).

Control of codling moth (*Cydia pomonella*) by „attract and kill" method

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The „attract and kill" method based on combination of attractant and insecticide was tested against codling moth from 2001 to 2004 in Prague – Ruzyně in experimental apple orchard. In 2001, the preparation Appeal (contained cyfluthrin 4% + codlemone 0,1%) produced by Bayer was tested on 0,34 ha plot. In 2002 - 2004 the preparation LastCall^{TM} CM (contained permethrin 6% + codlemone 0,16%) produced by IPM Technologies was tested on 1,7 ha plot. The efficacy of the tested preparations Appeal and LastCall^{TM} CM was ascertained according to level of fruit injury on experimental and untreated plot. In 2001, the fruit injury after the first generation was 15,8% on the plot treated by Appeal and 32,5% on untreated plot. The fruit injury at harvest was 6,4% and 26,3% on the plot treated by Appeal and untreated plot, respectively. In 2002, the fruit injury after the first generation was 1,1 %, on the plot treated by LastCall^{TM} CM and 19 % on untreated plot. The fruit injury at harvest was 0,2% on treated plot and 26,3% on the untreated plot. The results of 2004 will be presented.
This work was supported by the Ministry of Agriculture, the Czech Republic, project No. QD 1048.

Image Database File HANABACHI Based on the Japanese Bees

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An image database file HANABACHI (= bees in Japanese) is one of the files in KONCHU database treating species information on the Japanese, East Asian and Pacific insects. It is a species information database based on the Japanese bees including images. The HANABACHI contains image keys to the families and genera of the Japanese bees and species data. A user can also use related files, DJI (Dictionary of the Japanese Insects) and MOKUROKU (A Check List of the Japanese Insects) which are linked to the HANABACHI file. Each species record is composed of 18 items, such as a family, genus and species (or subspecies) names, Japanese name, distribution data, sex of the type, type locality, type depository, source of the original description, synonymies, morphological notes, remarks, flying season, visiting or associated flowers, parasites, references, various and distribution map. The image essentially includes the general habitus, head in frontal view, mesoscutum, propodeum and metasomal terga for each sex of the species. It is managed by a text database management system SIGMA. A user can access the database file by visiting the following URL: http://konchudb.agr.agr.kyushu-u.ac.jp/hanabachi/.

Invasive ants – knocking at New Zealand's door

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Within the last few years numerous invasive ants have been detected in New Zealand biosecurity border control areas -largely due to the instigation by MAF Biosecurity of a targeted ant surveillance programme at points of entry. These include *Solenopsis invicta*, *S. geminata*, *Paratrechina longicornis*, *Tapinoma melanocephalum* and *Anoplolepis gracilipes*. For some of the high-risk species New Zealand is too cold and they are unlikely to be significant pests. For others, like *S. invicta*, northern New Zealand is at risk. Why are these ants successful invaders? We look at their biological traits and New Zealand border interception data to highlight their success as stowaways, and also demonstrate the numerous entry pathways that exist for invasive ants. Recent changes to importation processes are aimed at picking up more stowaway organisms, and targeted ant surveillance will continue; however, with the ever-increasing importation of goods from around the globe and the continuing international spread of invasive ants, preventing future incursions will remain an ongoing challenge.

Use of mineral oil and predatory mites in the management of two-spotted mite in greenhouse roses

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Two-spotted mite, *Tetranychus urticae*, is an important cosmopolitan pest that causes severe economic damage in horticultural crops. Its resistance to many synthetic acaricides has resulted in the search for alternative control approaches. A series of experiments conducted in greenhouses on commercial rose crops investigated the effectiveness of mineral oil sprays and releases of the predatory mite *Phytoseiulus persimilis* in two-spotted mite integrated pest management programs. Application of 0.5% v/v sprays of an *n*C24 horticultural mineral oil applied fortnightly gave excellent protection from when two-spotted mite mite populations were below the economic threshold of 0.5 mites per leaflet before the first oil application. If oil was applied when mite populations were above the threshold, populations were stabilised but not reduced. Application of oil to the upper parts of canopy and release of predators improved control of two-spotted mite over the release of predatory mites alone. Comparisons of the cost of three pest management programs (oil alone; oil with *P. persimilis*; oil in rotation with synthetic acaricides) with the conventional synthetic acaricide program demonstrated cost savings of around 43%, 50% and 12% for these programs respectively.

Value of remnant native vegetation to horticulture

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There is an increasing emphasis on protection of remnant native vegetation and on constructing native vegetation shelterbelts and corridors to protect and manage biodiversity in our landscapes. This project is investigating opportunities to integrate natural resource management with arthropod pest control in irrigated horticulture in the Northern Mallee region. We also aim to increase community awareness of the role and value of native vegetation. We selected seven study sites where vineyards were adjacent to remnant native vegetation. At each site we established two transects perpendicular to the native vegetation/vineyard boundary and installed five pit-fall and five pan traps along each transect, to capture arthropods within the native vegetation and the vineyard. Traps are installed at the vineyard edge and 25 and 50 metres into the vineyard, and at the edge of the remnant native vegetation and 50 metres into the vegetation. We will collect samples from a one week trapping period four to five times a year. Samples already collected are being sorted to major groups of interest (Hymenoptera, Coleoptera, Diptera, Thysanoptera, Lepidoptera, Neuroptera, Dermaptera, and spiders, scorpions, centipedes and millipedes). Selected groups will be sorted to morpho-species to allow some analyses of diversity and functional groups.

Use of pheromones in integrated pest management in Uzbekistan

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The Institute of bioorganic chemistry of the Uzbek Academy of Science was founded in 1973. The Institute has developed new perspectives in the fields of bioorganic chemistry, molecular biology, biotechnology, and chemical ecology. A major research emphasis is structure-function studies of substances from animals and plants. We carry out investigation of pheromones of insect pests of agriculture in Uzbekistan. Particular attention is given to practical use of pheromones. Methods have been developed for synthesis and practical use of pheromones for more than 30 species of agricultural pests in Uzbekistan. The research includes synthesis of optically active forms of pheromones with chiral centers. Investigations are also conducted on the dependence of attractiveness on structure. The Institute of bioorganic chemistry produces and sells kits containing traps and pheromone lures. These are widely used in the agricultural areas in Uzbekistan, particularly in cotton. We investigate main cotton pests – *Heliotis armigera, Agrotis segetum, Spodoptera exiqua, Lygua lineolaris.* Pests of horticulture – *Euzophera punicaeela, Choreutis nemorana, Laspeyresia pomonella, Grapholitha molesta.* Pests of forestry – *Sarrothrypus musculana, Eurytoma samsonovi.* Pests of food-stuffs – *Ephestia kuehniella, Plodia unterpunctella, Sitophilus oryzae, Cryptolestes pusillus.* Pest of silk-tree – *Glyphodes pyloalis.*

Assessing the risk of reduced-risk apple pest control programs in New York State

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Apple is a high value crop in the US, worth US$1.5 billion annually, and US$100 million in New York State alone. A current on-farm project in NYS is concentrating on the use of IPM systems as an alternative to broad-spectrum insecticides that have been relied upon for 40 years, to greatly reduce residues and worker exposure to organophosphate, carbamate, and pyrethroid insecticides and to evaluate, on a regional scale, reduced-risk tactics that will be effective, sustainable, economically viable, and compatible with biological control. This study addresses the effectiveness of tactics that include IGRs, botanically derived and selective pesticides, and pheromone disruption. Results after 2 years of this 4-year study show that reduced-risk insecticides are effective against many secondary pests. However, fruit damage from internal lepidopterans and apple maggot in reduced-risk orchards tends to be slightly higher than that occurring in standard blocks. This project will help determine if selective insecticides, alone or integrated with mating disruption, can provide adequate control of direct pests of fruit for which there is no allowable market tolerance for damage. It will also help identify potential new pests, as well as natural enemies, that may occur in orchards treated with these new, selective tactics.

Vegetational diversity and conservation biological control

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The theory of multi-species interactions is rudimentary while vegetationally diverse habitats require examination of many interactions. We identify two 3-species interactions that may be crucial to understanding how natural enemies respond to vegetational diversity. We concentrate on cannibalism and intra-guild predation in a multi-predator system, and on the shared predator, apparent competition multi-prey system. *Coleomegilla maculata* [Coccinellidae] is cannibalistic and feeds on aphids, pollen and other species of coccinellids in maize fields. In one set of experiments, densities of *C. maculata* and other species of coccinellids were experimentally manipulated to evaluate the relative strengths of cannibalism and intra-guild predation and to predict if predation by the various species was redundant or complementary. By analyzing the system as a competitive system, we predicted that predation was complementary. There may be many circumstances that multi-predator systems simplify into ones structurally similar to two-species competition. In another set of experiments, densities of aphids, maize pollen and Lepidoptera eggs were manipulated experimentally to evaluate the effects of multiple prey on *C. maculata*. Unexpectedly, addition of aphid prey sometimes increased per capita predation rates on eggs. Careful examination of three-species interactions may reveal ways to integrate community interactions into a general theory of vegetational diversity.

Adoption of cotton IPM in the state of Andhra Pradesh, India

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Cotton is an important commercial crop of Andhra Pradesh state covering an area of around one million hectares accounting to 12.5 % of the state total cropping area during *Kharif* (rainy) season. The production of cotton is limited by insect pests to an extent of 50 % despite the use of around 45 % total pesticide consumption. Since 1940s, pesticide application has been the principal approach of plant protection. The pesticide based technology packages exacerbated the pest control problem by reducing pest defenders and development of insecticidal resistance. Hence, it is imperative that Integrated Pest Management (IPM) will play a crucial role in coming decades. *Has the theory of IPM delivered a change in practice?* Certainly yes, the period of transition from conventional paradigm of insecticidal control to pest management tools is underway. The present situation is such that both research and extension activities are to be weighed equally to achieve the production targets. The technologies to ameliorate cotton pest problems, pest monitoring and transfer of technology activities of the state agricultural university, activities of the state agricultural department such as dissemination of IPM package to clientele, season long training programmes on IPM, farmers field schools, establishment of biological control laboratories, encouraging *NGO's* participation, regulation of pesticide marketing and adoption of IPM at farmer level coupled with reduction in pesticide consumption over a decade were discussed. The need for establishing cotton clinics, development of curricula and training modules, information net work, educational software package on Diagnosis and Recommendation in most user-friendly version was emphasized.

Potential use of trap cropping to control diamondback moth in cabbage

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The ovipositional preference of the diamondback moth (DBM) *Plutella xylostella* (L.) (Lepidoptera: Plutellidae) and the larval survival of its larvae were tested on four hosts: cabbage, *Brassica oleracea* var. *capitata* L.; collards, *Brassica oleracea* var. *acephala* L.; Indian mustard, *Brassica juncea*; and yellow rocket, *Barbarea vulgaris* (R. Br.). These experiments showed yellow rocket and glossy collard as good potential trap crops. The percentage of trap crop plants necessary to make the trap crop was tested for these trap crops in six different treatments (0, 4, 8, 16, 24, and 32% of trap crop plants in relation to cabbage). Results of these studies and other ones aiming to determine factors of importance in the effectiveness of trap crops show that there is potential in the use of trap crops, particularly yellow rocket, to control diamondback moth in cabbage.

Mating disruption of several lepidopterous pests using high-emission-rate, widely spaced dispensers

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Widely spaced, high-emission-rate pheromone dispensers have been in commercial use for many years in both field and fruit-tree crops and allow a judicious use of pheromone while minimizing application costs for labor. This is the only type of mating disruptant formulation whose dispenser density (and amount of A.I.) per ha diminishes as the hectarage treated increases while maintaining the same dispenser grid-spacing, due to an advantage of geometry provided by widely spaced dispenser systems. The Metered Semiochemical Timed Release System ("MSTRS") evolved away from a clock-timer machine-driven 'active' release of pheromone into an entirely passive release system using polyethylene bags that maintained high-emission-rate low-point-source-density attributes. The application of this formulation against the European corn borer in Iowa reduced damage to corn by an average of 50% in replicated trials over several years. The density of bags needed to be deployed is only 25 bags per 10 hectares of corn. Merely delaying mating by females and not eliminating it entirely is responsible for the reduction in larval damage. MSTRS dispensers at a density of only 15 per hectare in cranberry marshes significantly reduced mating by free-flying black headed fire worm females. The system also is effective against the oriental fruit moth.

Population dynamics models of vegetation diversity and pesticide sprays in a broccoli-aphid agroecosystem: an alternative to ANOVA in gauging the effectiveness of IPM.

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We explored the interaction of vegetation diversity and selective pesticide disturbance on aphid population dynamics using an optimization approach that fits data from a factorial design field experiment to simple differential equation models. In particular, we established field plots of broccoli surrounded by (a) bare ground or (b) weedy margins, subjected to (i) no pesticide spray, (ii) low concentration pesticide spray, or (iii) high concentration pesticide spray. We fit data from weekly aphid censuses in treatment plots to exponential and logical growth models, and numerically solved an inverse problem in order to calculate time-varying birth/death rates for aphids in different treatment plots. The results of our optimization/inverse problem revealed a synergistic interaction between margin type and pesticide disturbance, corresponding to independent statistical analyses (MANOVA). This modeling approach provides a means of inferring information about mechanisms underlying population field data from factorial design experiments. Our results suggest that combinations of increased vegetation diversity and selective pesticides may act synergistically to reduce populations of target insect pests.

Biodiversity and parasitoid effectiveness: a new twist to an old story

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The direct positive relationship between biodiversity in agroecosystems and effectiveness of natural enemies has been an underlying assumption of agroecology. Yet recent research has suggested that although greater biodiversity may be necessary it may not be sufficient. That is, biodiversity may or may not enhance natural control depending on the nature of the biodiversity, e.g., what organisms constitute the diversity. We present the results of research that determined the traits that influence the susceptibility of macrolepidoptera to parasitism. We suggest that the extent to which species in a community exhibit traits enhancing susceptibility and the number of such species may determine if greater biodiversity leads to greater effectiveness of biological control agents.

Behavioural responses of arthropods to iso- and cycloalkanes and implications for use of horticultural and agricultural mineral oils

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Horticultural and agricultural mineral oils (petroleum spray oils) are important components of biorational integrated pest management programs. These products contain aromatics (0% to about 8% depending on oil quality), cycloalkanes and isoalkanes. Historically, emphasis has focused on their use in dilute (£ 2% v/v) aqueous emulsions to drown susceptible pests, mostly mites and scales. Recent studies have shown that a far wider range of pests can be controlled, with less oil, through the influences of spray deposits on feeding and/or oviposition behaviour. We review recent studies on tetranychid mites, thrips, whiteflies, Asiatic citrus psyllid, dacine fruit flies, *Helicoverpa* spp., agromyzid and gracillariid leafminers, and other pests, and discuss their relevance to pest and disease management programs. Possible reasons for the behavioural effects are also discussed. The effects do not depend on the aromatic content of oils. Queensland fruit fly and Oriental fruit fly respond, quite dramatically, to isoalkanes and cyloalkanes that boil within a very narrow range of temperatures. The results suggest that females perceive these molecules, or parts of them, to be naturally occurring molecules that are detrimental to the development of their larvae.

The influence of groundcover plant species on *Epiphyas postvittana* (Lepidoptera: Tortricidae) and its parasitoid *Trichogramma carverae* (Hymenoptera: Trichogrammatidae)

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Greenhouse experiments were conducted to investigate how *Trichogramma carverae* responded to different groundcover plant species. Adults (< 24h after eclosion) were caged with different groundcover plant species to measure longevity and daily fecundity. Both male and female longevity in *Fagopyrum esculentum* and *Lobularia maritima* treatments were significantly higher than on shoots of these species from which flowers had been removed. Daily fecundity was significantly greater in the *L. maritima* with-flowers treatment than in all other treatments. There was no significant enhancement of longevity or parasitism when wasps were caged with intact *Brassica juncea* or *Borago officinalis*. Separate greenhouse experiments quantified the effect of groundcover plant species on the pest, *Epiphyas postvittana*. Both adults and larvae benefited from *F. esculentum*. This result suggests that whilst *F. esculentum* is non-selective food plant, *L. maritima* could safely be used in a vineyard to enhance *T. carverae* with minimal risk of favouring the pest.

Western flower thrips pupation behaviour on greenhouse chrysanthemums and implications for integrated control

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Understanding the spatial distribution of thrips life stages on the host crop is essential for developing successful control strategies. Information is lacking on the pupation sites of western flower thrips (WFT), *Frankliniella occidentalis*, on different host plants. The proportions of WFT pupating on potted chrysanthemum plants and on the substrate were determined in two experiments (one in Canada and one in the UK). The experimental methods differed, but both included destructive plant assessments and counts of thrips larvae falling to sticky cards beneath the plants. In the Canadian experiment on cv. 'Miramar', 47% of thrips dropped off the plants to pupate and 10% of were found pupating on the plants. The remaining 43% were unaccounted for and presumed to be hidden within the flowers for pupation. WFT pupae were documented for the first time inside disc florets. In the UK experiment on cv. 'Swingtime', results indicated that 93-96% of thrips dropped to the ground to pupate and only 4-7% pupated on the plants. In this experiment, the few pupae found on the plants were primarily in open flowers including inside disc florets. The differences in the experimental methods and results and the implications for integrated control strategies will be discussed.

Development of IPM for thrips in greenhouses of chive.

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Chive *(Allium schoenopransum*; Liliaceae) is a major exporting crop for fresh herb growers in Israel. In recent years "silver spots", resulting from thrips feeding (both *Thrips tabaci* and *Frankliniella occidentalis*), significantly restricted the ability of local growers to export chive. We determined the distribution of these thrips outside and inside walk-in tunnels (10x30m) used for chive growing in Northern Israel. The study was conducted from September to July for three years. Thrips populations were highest during the Spring and Fall seasons. Outside the tunnels significantly higher numbers of thrips were trapped on the eastern side. About 75% of the thrips were caught between 7:00 to 13:00 and they were evenly dispersed in height between 0.5-2.0 meters. Inside the tunnels most thrips were caught at the south side, which is the hottest and has the most light. However, the highest numbers of thrips and damaged leaves were found on plants at the northwestern side which had the optimal climatic condition for their establishment. This information may be used to reduce the risk of thrips entry and establishment.

Genetic status of medfly in Australia as inferred from microsatellite markers

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The Mediterranean fruit fly *Ceratitis capitata* is one of the world's most important pest species. It is native of sub-saharan Africa, is highly polyphagous and has a long history of invasion success. In Australia, it was first captured near Perth in 1895 and soon spread throughout the country. As a consequence of the expansion of the native pest species, *Bactrocera tryoni*, medfly disappeared in East Australia by the 1930s, but resisted in the Western coastal region. The presence of an inhospitable land separating West and East Australia and the limited number of roads and rail routes, concurred in protecting South Australia from medfly infestations. However, during 2000/2002 several medfly outbreaks occurred nearby Adealide. With the aim of deriving a general picture of the status of medfly in Australia and detecting the possible origin of medfly bioinfestations in South Australia, the genetic status of medflies from South and West Australia was analysed at ten SSR loci and compared to that of flies from Africa, south Europe and Hawaii. The combination of phylogeographic analysis and assignment test approaches showed that overall Australian medflies are most similar to Mediterranean medflies and that flies from South Australia are most probably derived from West Australia.

Common salt as a mirid management tool

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Common salt (NaCl) at 0.5% (5g/L of water) gave significant control of mirids (predominantly *Creontiades dilutus* (Stål)) in two trials in podding mungbeans (*Vigna radiata*). In the first trial at 3 day after treatment (3 DAT), salt gave 80% control of adults, but only 28% control of nymphs. In the second trail, salt gave 70% and 77% control of nymphs and adults at 1 DAT respectively, and 74% and 56% control respectively at 4 DAT. However salt had little residual activity, giving poor mirid control by 9 and 7 DAT in trials 1 and 2 respectively. The addition of salt (0.5%) significantly improved the effectiveness of low rates (£ 250 mL/ha) of dimethoate (Ȯ at 500 mL/ha) against mirids. In two trials, dimethoate plus salt treatments had on average only half as many mirids as the corresponding rates of dimethoate without salt. While dimethoate, plus salt, at rates as low as 125 mL/ha, gave excellent control of mirids present at spraying for 2-4 DAT, dimethoate plus salt at < 250 mL/ha gave poor control of nymphs hatching (from eggs) post spray. Populations of small nymphs reached pre-spray levels by 6 DAT for the lowest dimethoate rate trialed (62.5 mL/ha).

Failure of mating disruption for *Helicoverpa armigera* H. (Lepidoptera: Nocutidae) in isolated tomato and capsicum crops.

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A full scale trial of mating disruption for *Helicoverpa armigera* was run in isolated tomato and capsicum crops in the Promised Land cropping region, south-eastern Queensland. The cropping area in this region consists predominantly of a mosaic of tomato, capsicum, sugarcane, citrus and avocado. The region is isolated from sources of *H. armigera* adults by at least 6km of state forest which is virtually devoid of larval host plants. We attempted to prevent *H. armigera* mating by treating all solanaceous crops within the region with mating disruption dispensers. Monitoring with pheromone traps and sentinel females indicated that complete mating disruption was achieved in the treated areas, but there was no corresponding decrease in the number of eggs laid in the treated crops, or in the proportion of mated wild females captured in light traps compared with untreated regions. Further trapping and observations indicate that mating was occurring on untreated non-host plants such as sugar cane adjacent to the treated areas. This is a novel finding and contradicts published observations and assumptions made in regard to pheromone production and mating behaviour in heliothine moths. A commercially viable mating disruption program in the Promised Land region would require treating both host and non-host plants within the cropping region. This is currently neither economically nor logistically feasible.

IPM of protected cropping in established northern greenhouse regions

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Biological control of pests in Northern Europe and Canada was taken up in the late 1970s in vegetable crops due to severe problems with pesticide resistance in glasshouse whiteflies and two spotted spider mites. In the Netherlands, the UK, and Scandinavia biological control of the two pests soon proved to be viable and economically feasible and the method was almost immediately implemented by the glasshouse industry. Besides growers' positive attitude, the climate and glasshouse constructions have facilitated the effect of biological control agents. A key aspect is that in the major part of the growing season, the glasshouses can be considered as closed environments with little influx of pests. Another aspect is the generally high sanitary standards of the glasshouse operations in these regions that reduce refuges of pests and thereby increase the control effect on the crop. The research community has focused on developing biological control agents for former secondary pests and the commercial producers have been very effective in utilizing the scientific results. Insectaries have, thereby, been able to provide biological control solutions to pest problems soon after they have emerged in both vegetable crops and ornamentals. Until recently, a rather liberal legislation on importation of exotic beneficial arthropods into continental Europe has facilitated the utilization of the same biological control agents across borders. This increases the market for insectaries and the availability of biological control agents to growers.

Western flower thrips: Australia's unwanted immigrant

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Western Flower Thrips, *Frankliniella occidentalis*, is a recent introduction to Australia. WFT were first detected in Perth in 1993. They have since spread to all states of Australia, with the exception of the Northern Territory. WFT cause damage to crops through direct feeding, and through virus transmission. Wide ranges of crops are affected including vegetables, ornamentals and fruit crops. Insecticides are favoured by commercial growers in Australia as their main control method. However, as has occurred elsewhere, WFT are resistant to a number of insecticides. Furthermore, the current registered rate of many insecticides in Australia may be too low. This would increase selection for resistance, because pesticides are being applied at a sub-lethal dose. Resistance has already been detected against the newer chemistries such as spinosad and fipronil, albeit at a low level. In the long-term, insecticidal control of WFT is unsustainable. An integrated approach must be adopted. However, few other control methods are available to Australian growers. For example, biocontrol agents effective overseas are unlikely to be approved for release here. This is due to Australia's quarantine regulations that govern the importation, testing and release of exotics. Nevertheless, other control methods are required.

Pheromone based mating disruption strategy in agroforestry.

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Western poplar clearwing moth (*Paranthrene robinae* Hy. Edwards) populations have reached outbreak proportions in hybrid poplars grown for pulp and saw timber in eastern Washington and Oregon. Not even recorded as a serious pest of poplar in 1999, there were only 95 *P. robinae* males captured in 2001, but the following year the same number of pheromone baited traps captured 17,000 male moths. Clearwing larvae burrow into the heartwood of these trees. Repeated applications of chlorpyrifos in 2002 had little or no effect on the number of males captured in baited traps or damage to trees. We treated 3,823 Ha with synthesized female pheromone [4:1 ratio (E,Z):(Z,Z)-3-13-octadecadienyl alcohols] beginning June 2, 2003. Formulations included membrane dispensers and aerial applied flowable microcapsules (WPCM-F Suterra®, Bend OR). After pheromone application the trap capture was reduced by a dramatic 99.75%; however, some damage was attributed to moths that had emerged and mated before the June application. Application in 2004, started in April and the entire 14,164 Ha was treated with pheromone. This is the first attempt to use a pheromone based 'mating disruption' strategy on a large scale Agroforestry system.

Disruption of moth mating with pheromones: formulation and behavior determine efficacy

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Worldwide some 430,000 ha of crops and forests are protected from many moth pests by application of formulated pheromone to disrupt mating, typically as component of a management program that also includes application of conventional pesticides. The pests controlled span a spectrum of moth families, pheromone chemistries, and crops infested. The methods of dispensing pheromone varies widely, from sprayable microdispersibles, to point sources that mimic the release characteristics of an individual female, to point sources emitting pheromone at rates many thousands of times the release rates of females. The most commonly invoked explanations of mating disruption invoke adaptation of peripheral sensory receptors, CNS habituation of responsiveness, competition between pheromone sources and females, and camouflaging of the female's plume. The type of formulation, the nature of the plant canopy, wind conditions, and the sensory and orientation capabilities of the particular moth species, together dictate which mechanism predominates in achieving disruption of mate finding. But most successful cases of disruption seem to hinge on multiple, interacting mechanisms. Understanding how these can be optimized in a formulation for a given species may allow improvements in efficacy.

Development of integrated fruit production for apples in Eastern Canada: making use of pesticide risk indicators to facilitate adoption of IPM practices and to monitor changes in pesticide use over years

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In North America as in most other regions of the world, numerous policies aiming to reduce the use of pesticides were implemented in the past 20 years with variable success. Among encountered problems, the debatable choice of an indicator to measure the progress of these programs can be pointed out. We used data collected since 1977 in a series of apple orchards in Quebec, Canada as an example to demonstrate a decline of the environmental impact of spray programs used in this region against insects and mites. During this period, the average field value of the Environmental Impact Quotient (EIQ) decreased by 66%. The same decrease was not always noticed, however, when other indicators were used, such as the number of applications or the amount of active ingredient used per hectare. Since the use of precise quantitative tools can make a difference when drawing conclusions from pesticide use data, we suggest to make use of EIQ or similar risk indicators to facilitate the classification of pesticide applications according to recommendations in IFP programs currently under development for apples grown in Canada.

When can pestiferous mirid bugs be beneficial biological control agents?

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Insect ecologists have only recently begun to acknowledge that many species exhibit omnivorous feeding habits as they exploit both plant-based resources and arthropod prey. The prevalence of omnivores in agroecosystems calls for special attention in IPM programs. Understanding the feeding habits of omnivorous pests is particularly important for mirid bugs, as several economically important pests in this family also feed on other arthropod pests in the system. It appears that mirid populations previously suppressed by insecticides, tend to build up under IPM regime. A better knowledge of their feeding biology will thus help assess their overall economic impact and allow the development of new approaches to population management. The major challenge facing applied entomologists is to identify the conditions, including plant characteristics and availability of arthropod prey, which may influence levels of omnivore-inflicted crop damage. This is the objective of our research-in-progress on *Creontiades pallidus* in greenhouse sweet pepper. Results to date indicate that different prey species in the system, such as red spider mite, sweet potato whitefly, western flower thrips, and moth eggs, differ in their nutritional suitability for the bug. I will discuss the relationship between prey availability and *C. pallidus*-inflicted damage to sweet pepper fruits.

Attraction of male *Lygus rugulipennis* to components of the female sex pheromone

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We previously showed that three chemicals, hexyl butyrate, (*E*)-2-hexenyl butyrate and (*E*)-4-oxo-2-hexenal are present in both male and female adults of the European tarnished plant bug, *Lygus rugulipennis* (Poppius) (Heteroptera: Miridae), but that only the females produce these in detectable amounts when undisturbed. They were suspected to be components of the female sex pheromone, but field trapping experiments using polyethylene vials and sachets as dispensers were unsuccessful. Here we report more recent field experiments during which the chemical components were released from microcapillary tubes. The mixture of hexyl butyrate and (*E*)-4-oxo-2-hexenal was significantly attractive to male *L. rugulipennis,* and a blend of all three components attracted significant numbers of males of the congeneric species *L. pratensis*. The possible reasons for the success of the microcapillaries compared to other dispensers are discussed in terms of the release rate, the release ratio and the importance of a point source dispenser.

Pushing insects around: the use of 'push-pull' strategies in integrated pest management

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'Push-Pull' strategies could further decrease pesticide usage in IPM systems by reducing insect pest infestations through the use of repellents / deterrents to 'push' pests from the main crop while simultaneously attracting or 'pulling' them onto neighbouring, more preferred host plants (trap crops), where they can be more efficiently controlled. We explain the principles of push-pull strategies and demonstrate their potential using a model system. Oilseed rape (*Brassica napus*) can be protected from damage by the pollen beetle (*Meligethes aeneus*) through use of Lavender essential oil and turnip rape (*Brassica rapa*). Lavender oil shows potential as an effective repellent; it deters pollen beetles from colonising treated oilseed rape plants. Turnip rape acts as an efficient trap crop; pollen beetles prefer turnip rape to oilseed rape. The mechanisms of attraction were investigated to maximise the efficiency of this component of the strategy. In the field, a border of turnip rape surrounding oilseed rape plots reduced infestation of the oilseed rape to below spray-threshold levels, and damage was significantly lower than in control plots. Turnip rape can also augment biological control of the pollen beetles by positively influencing the distribution of their parasitiods.

Grape IPM and biological control in California organic vineyards

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Development of novel approaches for Integrated Pest Management (IPM) systems has often relied upon research in organic or sustainable crops systems. California vineyards were at the forefront of many novel approaches developed in the 1950-70s, with vineyard insect pests the target of classical, augmentation, and conservation biological control, as well as many cultural control practices. Today, the practice of organic vineyard farming in California is jeopardized by a series of newly invasive pests, such as the glassy-winged sharpshooter and the vine mealybug. New control options currently developed for these pests include biological control, the use of area-wide management (glassy-winged sharpshooter) and mating disruption (vine mealybug). Nevertheless, these approaches have yet to be proven effective for organic producers that have infestations of these newly invasive insect pests. As a result, successful organic farming may rely more on pest species present, vineyard location and commodity (wine, raisin or table grape) than on implementation of sustainable insect management practices. I discuss both past and current insect pest management programs, as well as the outlook for organic farming in California vineyards.

Plant extracts as source of semiochemicals and fumigants for thrips control

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Many plant species have insect repellent or pesticidal properties. Extracts from these plants can be used to develop pesticides or repellents to be used in pest control strategies. We have selected several (compounds from) plant extracts that show in laboratory assays repellent and/or toxic effects on several species of thrips: *Frankliniella occidentalis* (western flower thrips), *Thips tabaci* (onion thrips) and *Thrips simplex* (gladiolus thrips). Current research focuses on the development of control strategies for thrips species based on these compounds. These strategies include: 1) repellents as synergist of pesticides, 2) plant extracts as fumigants for disinfection, 3) formulated extracts as toxic and/or repellent crop spray to keep thrips away. We will present examples from these strategies that show that plant extracts have potential for control of thrips.

Effective emulsified wax dispensers for pheromone-based pest control

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Atterholt (1996) demonstrated the potential of using emulsified paraffin wax to release insect sex pheromones, creating biodegradable Emulsified Wax Dispensers (EWDs) which are more economical, adaptable, and easier to apply than other available pheromone dispensers. Optimization of EWDs at Michigan State University, U.S.A., for mating disruption of *Grapholita molesta* resulted in the development and field-testing of a dollop formulation that was as effective as the commercial standard (Isomate M-100 ropes, Shin-Etsu Chemical Co., Ltd.) season-long in peaches (*ca.* 15 wk) and could be applied almost twice as fast. In subsequent studies, substituting microcrystalline wax for paraffin improved the formulation by reducing the emission rate of *G. molesta* pheromone by half and increasing dollop adherence to tree bark. ISCA Technologies, Inc. (Riverside CA, U.S.A.) scaled-up production of the microcrystalline formulation. This commercially-available Semiochemical and Pheromone Lure Application Technology (SPLAT) is providing excellent protection against *G. molesta* through mating disruption and attract & kill in large-scale commercial enterprises. By reducing the cost of pheromone-based pest control, SPLAT should encourage wider adoption of this technique in organic and IPM programs worldwide. Atterholt, C.A. 1996. Controlled release of insect sex pheromones from sprayable, biodegradable materials for mating disruption. Ph.D. dissertation. University of California, Davis, CA.

Organic pest management in the U.S. grain belt: challenges and perspectives of an increasing trend

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Organic soybean [*Glycine max* (L.)] production in the U.S. has risen to more than 60,000 ha in less than 10 yr. and other organic grain crops have also experienced tremendous growth. This presentation reviews current organic pest management research and producer findings across the Grain Belt. Critical challenges associated with organic soybean production include soybean aphid (*Aphis glycines*), other seed-staining fungi such as *Cercospora kikuchii* and *Fusarium*, and bean leaf beetle (*Ceratoma trifurcata*) as a vector for bean pod mottle virus. Despite challenges facing producers from these insect pests, an economic evaluation based on three years of research at the Neely-Kinyon Long-Term Agroecological Research site near Greenfield, IA revealed that food-grade organic soybean could return \$1160 to 1250 ha^{-1} compared to \$235 for conventional soybean grown in a 2-yr rotation with corn (*Zea mays* L.). At the Neely-Kinyon site, staining ranged from 7.5 to 22% while yields ranged from 3.5 to 3.8 Mg ha^{-1} in 2002. Other IPM successes include cucumber beetle and squash bug in organic squash, and codling moth management with mating disruption and approved organic treatments. Despite the many challenges, increasing consumer demand for certified organic crops warrants more aggressive and participatory IPM research around the world.

Developments in organic IPM research and priorities for the future

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Organic agricultural research in the United States is undergoing a rebirth of interest, focus, and scope involving federal, state, university, and private sector partnerships. Still, less than 0.2% of the available research acres at academic institutions (US Land Grants) are devoted to organic research. Increased numbers of competitive funding opportunities exist from federal (USDA), state universities, state departments of agriculture, and the non-profit sector. The USDA-CSREES-SARE program, the USDA Organic Transitions Program, and The Organic Farming Research Foundation provide visible and targeted funding for organic agriculture research. Consumers, farmer groups, local non-profit organizations, and selected academic institutions are providing increased visibility, accountability, and prioritization to needed organic research and policy. A unique partnership of farmers and scientists (SCOAR) has been organized to develop a prioritized research agenda for pursuit of multi-disciplinary research and extension on working farms. US Midwestern farmers rated weed management, crop rotations for pest management, cover cropping, and the relationship of organic systems to product nutritional value as high research priorities.

Host plant-derived volatiles attract female oriental fruit moth *Cydia (Grapholita) molesta* in the laboratory

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The oriental fruit moth, a key pest of peach, also caused considerable damage to apple in recent years, particularly in the later part of the growing season. A small proportion of the population has the capacity for long distance flight and gravid females appear to be the main dispersers. With the ultimate goal of developing a monitoring system for females, peach shoot volatiles were analysed and tested for behavioural response of female moths. The following results and conclusions were made: (1) Mated female moths were attracted to volatiles from peach shoots, even when visual contact was completely prevented. (2) 22 compounds were identified and quantified from peach shoot, using direct thermodesorption followed by coupled gaschromatography-mass spectrometry. (3) The bioactive constituents were the short-chain compounds as found by bioassay-assisted fractionation based on selective sorbent polymers ("subtraction approach"). (4) A three-component mixture originating from two different metabolic pathways showed significant bioactivity. This synthetic mixture elicited a similar attractant effect for female oriental fruit moths as the full natural blend from peach shoots and the bioactive fraction. Further work is in progress to develop a surveillance system for use under field conditions.

Host switching during the season: sex-dependent flight behaviour and monitoring in a lepidopteran pest, *Cydia (Grapholita) molesta*

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Inter-orchard flight of moths may have important consequences for pest management in orchards, as exemplified by *Cydia (Grapholita) molesta*. Originally known as a key pest of peach, it has been found recently in several countries to cause significant damage to pome fruit orchards in the later part of the season. Mechanisms underlying this late-season host shift have been investigated using behavioural assays in a dual choice odour arena as well as using comparative assessments on a computer-linked flight mill, with the following results and conclusions: (1) There is no indication that olfactory cues emanating from pome compared to peach fruit trees elicit any preference in the moths that could promote a host shift. (2) A proportion of the moth population is capable of undertaking long flights, with gravid females being the main dispersers. (3) Changing abiotic but not changing biotic conditions during the insect's development were found to correlate with markedly increased flight in the adult stage. The most powerful influence was noted for decreasing photoperiod at constantly high temperature during the preimaginal phase. It is postulated that decreasing photoperiod during the insect's development is the main factor eliciting the late summer host shift observed in this lepidopteran pest.

Areawide organic IPM in pear: a 3-year study

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Areawide management programs for insect pests of apple and pear in the Western US have been successful since their inception a decade ago, targeting codling moth through mating disruption to replace organophosphate insecticides. Pear psylla, another important pear pest, is also appropriate for areawide management, being highly dispersive with potential natural enemies in surrounding habitat. Establishing organic orchards among conventional orchards has been difficult: pests readily migrate from adjacent conventional orchards, yet natural enemy movement is limited by the pesticide barriers. Organic IPM on an areawide basis provides opportunities for immigration of biocontrol agents. In 2002, an Areawide Organic Management Program was established on 310 ac of contiguous pear, surrounded by native vegetation. Organic IPM practices were implemented for insect control throughout the project. However, other organic practices were not required (e.g., nutrient, rodent, and weed management), and approximately 50% of the acreage was Certified Organic. Over three years, there was a reduction in pesticide use and insecticide costs, with no differences in pest densities. However, there have been no correlated increases in natural enemy densities. Fruit yield and quality have been maintained, and alternative marketing programs have been attempted.

Habitat manipulation – half a century in waiting

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By the 1950s, the components of habitat manipulation that we use today to favour natural enemies were already established - nectar feeding, alternative and alternate hosts and microclimate. However, neither farmers, politicians nor the public were interested. The newly-arrived organochlorine insecticides were cheap, efficicnt and as yet irreproachable, biocontrol was only about importation, no one wanted weeds on a farm and diversity was an arcane and complex concept. What a 'sea-change' there has been! Environmentalists have fuelled an unreasonable public distaste of insecticides, that same public now demands cheap food from a beautiful and diverse agricultural landscape, and we in Europe have realised that our despised indigenous natural enemies have, already for a long time, been seen in the New World as valuable imports for biocontrol.. The principles of the 50s have now become farmer practice in Europe. Much of the purposeful introduction of 'biodiversity', however, is not designed for the improved biocontrol it achieves as much as for survival of 'our feathered friends'. Economically viable options for habitat manipulation will be described.

Plant bug (*Lygus lineolaris*, *Lygocoris inconspicuous*, and *Taedia scrupeus*) biology on strawberries and grapes

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Plant bugs are a common constituent of the arthropod communities of cultivated grapes and strawberries in the Northeastern USA. *Lygus lineolaris* (Tarnished Plant Bug or TPB) is a well-recognized direct pest of strawberries while *Taedia scrupeus* (Banded Grape Bug or BGB) and *Lygocoris inconspicuous* were thought to have little impact on grapes. We initiated research on ecology of all three species with different goals in mind. For TPB, we examined use of reflective mulch, variation in host plant resistance/tolerance, and influence of host plant density/patch size on abundance and damage to develop insights into non pesticidal approaches for control. Reflective mulch surrounding strawberry plants resulted in increased plant productivity, reduced nymphal densities, and reduced damage. We found evidence of cultivar differences in TPB damage mediated through multiple mechanisms but predominantly from increased productivity. Higher density of fruit was associated with less injury per fruit after controlling for TPB density indicating a saturation functional response. In grapes, we found that nymphs of BGB and *L. inconspicuous* are capable of causing significant reduction in yield at low densities (1 nymph per 10 shoots) mainly through reduction in number of clusters. Monitoring guidelines and control recommendations are in development.

Suppression of caterpillar pests in cole crops using pheromone-based attracticides

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Cabbage looper (*Trichopulsia ni*), diamondback moth (*Plutella xylostella*), and imported cabbageworm (*Pieris rapae*) are among the most serious pests of cole crops in the United States. The objective of this research is to determine the feasibility of using sex pheromone-based attracticide formulations to control these lepidopteran pests in cole crops. We evaluated two sex pheromone-based *LastCall*™ (attracticide) formulations (IPM Tech. Inc., Portland, Oregon, USA): one for *P. xylostella* and one for *T. ni.* Laboratory tests showed that male moths of each species were able to locate and make contact with conspecific *LastCall*™ droplets, and that minimal contact with *LastCall*™ resulted in significant moth mortality. In field evaluations, we recorded suppression of trap catch, and significant reductions in larval infestations and crop damage in *LastCall*™-treated plots, compared to untreated plots. Damage ratings for cabbage heads and collards in *LastCall*™-treated plots were similar to ratings in *Bt* plots, and both were significantly lower than in control plots. These results suggest that applications of sex pheromone-based *LastCall*™ formulations for *P. xylostella* and *T. ni* can provide control levels comparable to *Bt*.

Evaluation of conservation strips as a conservation biological control technique on golf courses

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Conservation strips combine the conservation biological control tactics of beetle banks and flowering insectary strips to create aesthetically pleasing refuges for predatory arthropods. The conservation strips contained two flowering plants, alyssum and coreopsis, and ornamental switchgrass. Conservation strips were established next to golf course fairways to evaluate their effect on arthropod abundance and distribution. Spiders, carabid beetles, and staphylinid beetles were of particular interest. They are the most common predators on golf courses and have been shown to inhabit beetle banks. Conservation strips supported a greater abundance of predators, parasitoids, and alternative prey than turf in the fairways or roughs. In addition, fairways and roughs adjacent to conservation strips had a greater abundance of predators and alternative prey than fairways and roughs without conservation strips. Black cutworm, *Agrotis ipsilon* (Lepidoptera: Noctuidae), is a common pest of golf course fairways. Predation of black cutworm larvae was significantly more frequent in fairways when conservation strips were present. For these reasons conservation strips show potential as a conservation biological control tactic on golf courses and agricultural systems. Manipulation of golf course habitat, by installing conservation strips, could reduce pest pressure and the need for insecticide applications.

Pest management in Australian sorghum – a success story

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During the past decade there have been dramatic changes in the management of the two main pests of Australian sorghum, the sorghum midge, *Stenodiplosis sorghicola,* and the corn earworm, *Helicoverpa armigera.* More than 95% of sorghum crops now have resistance to sorghum midge. Levels of resistance range from x2 to x12. Insecticide sprays to control midge have reduced from about 20-30% of crops being sprayed to less than 1%. The corn earworm has developed resistance to most of the synthetic insecticides previously used. A *Helicoverpa* specific nucleopolyhedrovirus (NPV), is now widely used with a concurrent marked decline in the use of broad-spectrum chemical insecticides. Currently, more than 95% of the treated crop is sprayed with NPV. Minimal spraying of sorghum crops with hard insecticides has increased survival of natural enemies and changed sorghum from a 'source' to a 'sink' for corn earworm, and a nursery for beneficial insects, including *Trichogramma, Microplitis,* Coccinellidae and Syrphidae. Sorghum's new attributes are being exploited in area-wide management strategies for corn earworm. A combination of plant resistance for midge and NPV/natural enemies for corn earworm has revolutionised sorghum pest management in Australia.

Citrus leafhopper IPM in Queensland, Australia

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Empoasca smithi, is a serious pest of the valuable early varieties Imperial mandarin and Navel orange, and in bad years also of mid- and late-season varieties in Queensland's Central Burnett area. Adults, which breed and feed on flush growth through spring and summer, switch in autumn and winter to green mature and colouring fruit where they cause skin blemishes, downgrading of up to 80% of fruit. Two mymarid wasp parasitoids, *Stethynium empoascae* and *Anagrus baeri*, kill up to 100% of eggs, but need insecticide assistance in most years. Endosulfan at 30 ml/100 L is cheap and effective, killing both nymphs and adults, but must be re-applied to kill hatching nymphs. The insect growth regulator buprofezin kills only nymphs, but also is very effective, though expensive - but contributes significantly to red scale and mealybug control. An orchard-wide strategy and good scouting and decision making is required to prevent re-infestation of treated trees by adults dispersing from hardening flush in untreated trees, especially in late autumn from mid-season varieties with non-susceptible fruit to early season varieties with maturing fruit, and from the common exotic weed tree species *Celtis sinesis*. The benefit to growers of this research is estimated at $11,250-$38,500/ha.

Greenhouse IPM - a part of a holistic integrated crop management approach

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Greenhouse production is a complex undertaking, comprised of several disciplines, among them entomology, agronomy, pathology, engineering, marketing and economics. Greenhouses are controlled environment structures considered to alleviate or eliminate uncertainty due to fluctuation and extremes in weather characteristic of field production. However, having this added control over plant production growers face the challenge of setting priorities regarding crop production components given that practices in one system can affect the outcome of management practices in another area of crop management. In addition to plant and pest management, a grower must make decisions regarding structures, staffing, scheduling, markets, and depending on locality or size of operation, export restrictions and international treaties. Integrating all these aspects of crop management to obtain optimum yields and profitability is a difficult task. A description of how integrated pest management practices impact and are impacted by crop production systems and how best to integrate these disciplines will be provided.

IPM in field crops

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Four species of thrips are common in northern Florida. Three are vectors of tomato spotted wilt virus. The population dynamics of individual species varies in different crops. Tomato is a poor reproductive host for each species. Pepper is a food and reproductive host for *Frankliniella occidentalis*, *Frankliniella tritici*, and *Frankliniella bispinosa*. Peanut is a food and reproductive host for *Frankliniella fusca*. This talk will present IPM programs for thrips and tomato spotted wilt in these crops. The IPM programs were developed from knowledge of the population dynamics of thrips and their role in the spread of tomato spotted wilt virus. The predator *Orius insidiosus* is an effective natural enemy in pepper resulting in suppression of thrips populations. Other tactics provide additional control including reduced-risk insecticides that conserve populations of the predator. An integration of UV reflective mulch, reduced-risk insecticides, and an acquired systemic resistance inducer provide control of thrips in tomato, and they reduce primary and secondary spread of tomato spotted wilt virus. A natural enemy *Thripinema fuscum* suppresses populations of *Frankliniella fusca* in peanuts and it is effective in preventing cycles of secondary spread. This natural enemy is compatible with the use of cultural and insecticidal tactics.

Disruptive factors hindering IPM implementation in protected cropping

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In Europe and Canada, IPM in the protected cropping industry, particularly in greenhouse vegetable crops, has achieved success and is now regarded as standard practice. Despite this situation, IPM programs continue to evolve with the development of new biocontrol agents in established greenhouse regions and the introduction of IPM programs in developing greenhouse regions. Additional external factors have the potential to disrupt these efforts. Factors such as pesticide toxicity, lack of IPM-compatible chemicals, new invasive pest species, regulations for registration of biocontrol agents (national and global), export rules (zero tolerance), consumer perceptions/expectations and lack of education, and technology transfer are discussed.

Commercial-scale field trials of attractants for *Helicoverpa* moths based on plant volatiles

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Plants emit volatile compounds that attract insects for foraging and oviposition. We have developed plant volatile-based attractants for *Helicoverpa* moths that can be used for attract-and-kill in cotton and other crops like corn and legumes. These formulations are mixed with an insecticide, methomyl, and sprayed on crops. Typically one row in every 72 is treated. In the 2002/03 season, two fields of cotton, approximately 40 ha each, were treated three times. The impact on treated and surrounding untreated fields was monitored by counting and dissecting dead moths to estimate the number of eggs which were not laid as a result of the treatment, and by regular scouting for a range of pest and beneficial insects. In the 2003/04 season, approximately 13,000 ha of cotton were treated under a product evaluation permit from the Australian Pesticides and Veterinary Medicines Authority. Results from both seasons indicate that attract-and-kill for female *Helicoverpa* spp. using plant volatiles could make a significant contribution to IPM on an area-wide basis.

Efficiency of parasitoids from a behavioural perspective

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Whilst parasitoids benefit from host herbivore-induced plant volatiles, their behavioural response to these olfactory cues is variable. This variability can cause the inconsistent performance of parasitoids as natural antagonists, but it may also facilitate improvement of their efficiency in biological control through genetic manipulation. Using a model system consisting of cabbage plant *Brassica oleracea*, herbivorous insect *Pieris brassicae*, and parasitic wasp *Cotesia glomerata*, we address how genotypic differences in olfactory response influence host parasitism, and how genetic manipulation of the odour-guided behaviour can enhance the efficiency of parasitoids. Bidirectional selection resulted in two genetically diverged strains of *C. glomerata* in behavioural response to the host-induced plant volatile. Simulation experiments demonstrated that the capacity of both habitat colonization and parasitism significantly increased in the high olfactory response strain, as compared to the low olfactory response strain. Thus, the efficiency of parasitoids can be improved by genetic manipulation of relevant traits. A newly developed technique to mark parasitoids with a stable isotope makes it feasible to test this postulate in the field. The novelty of this technique allows the movement of parasitoids and the resultant parasitism to be tracked at a spatial scale.

New frontiers in habitat manipulation: a 'reductionist' versus 'biodiversity' schism?

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This paper will explore an emerging tension between a 'reductionist-paradigm' and a 'biodiversity-paradigm'. In the former, pest management scientists typically apply quantitative criteria to the selection of habitat manipulation strategies. Examples from Australian work will illustrate the utility of this approach for the enhancement of parasitoids such as *Trissolcus basalis*, an important stinkbug natural enemy. 'Reductionist-paradigm' strategies offer scope for incorporation into high-input, arable crop production systems with low risk of adverse consequences and high likelihood of acceptance by farmers. The 'biodiversity-paradigm' for habitat manipulation is less concerned with the integration of carefully selected species into crop systems but focuses on the net benefit of the ecosystem services that are provided by species-rich communities such as weed strips or patches regenerated woodland. Such approaches to habitat manipulation may more risky in terms of pest management and less likely to be accepted by many types of farmer but potentially offer scope for a wider range of ecosystem services. Accordingly, they are increasingly promoted for reasons such as wildlife conservation and hydrology. Pest managers are developing an understanding of the benefits to them of farmland biodiversity, as will be illustrated by current Australian work on floristically diverse shelterbelts.

Feeding behavior of *Lygus hesperus* and *Lygus lineolaris* on whitefly life stages

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Although omnivorous *Lygus* spp. feed on and cause economic damage to a wide variety of important crops, the degree of their predatory activity is unknown. A study was conducted to compare the degree of herbivory and carnivory between *Lygus hesperus* and *L. lineolaris*. The feeding behavior of the 3rd, 4th and 5th instar and adult *L. hesperus* and *L. lineolaris* was quantified in feeding arenas containing a surfeit of *Bemisia tabaci* eggs, nymphs and adults on a cotton leaf. *Lygus lineolaris* generally fed more frequently and for longer periods of time on the cotton leaf than *L. hesperus*. Both *Lygus* spp. exhibited similar patterns of prey choice with a preference for whitefly nymphs > adults > eggs. *Lygus hesperus* consistently consumed more of each whitefly lifestage than *L. lineolaris*. Moreover, the amount of time spent feeding on whitefly nymphs was significantly less for *L. hesperus* than for *L. lineolaris*. The data obtained from this study suggest that *L. hesperus* is a slightly better adapted predator than *L. lineoloris*.

Evaluation of mating disruption for the control of the jasmine moth, *Palpita unionalis* (Lepidoptera: Pyralidae) in Egypt

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In olive groves the jasmine moth, *Palpita unionalis* (Hübner), maintain low to moderate population densities, however periodically outbreaks occur causing severe damage to olive trees. Control measures are based on conventional pesticides. The use of pesticides has caused serious problems: environmental pollution, development of insecticide resistance. This has caused concern and has led to a concerted effort for the development of Integrated Pest Management (IPM) systems. Mating disruption with synthetic sex pheromones is a promising method to be used in IPM. The effect of mating disruption on the population of *P. unionalis* was evaluated for two successive years of 2002 & 2003 in Egypt A formulation of the two female pheromone components, E11-hexadecenyl acetate and E11-hexadecenal (E11-16:AC/E11-16:Ald), at a ratio of 7:3 in Montmorillonite K-10 was prepared. Measurements of the pheromone release rate indicated high emission of E11-16:Ald, 80% percent of the aldehyde was released within two weeks following installation. The emission rate of E11-16:AC was lower, more than 50% of the initial dose used was present 6 weeks after exposure to the field. Results of large-scale field trials in a highly infested olive grove near Cairo/Egypt revealed male disorientation around 90-95%. Female egg laying and tree infestation levels were reduced on the treated site in comparison to the control.

Mass trapping of the olive moths

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A simple and inexpensive UV-trap was developed by the first author from materials generally available around the olive farm, to capture the main lepidopterous pests of olive trees. The trap consisted of a sticky-collecting device, two ultraviolet fluorescent tubes and tilted mirrors to increase light intensity by interference of reflected rays and spreading light reflection over a wider area. The trap was compared with delta-traps (baited with (Z)-Z-tetradecenol) and funnel traps (baited with (E)-11-hexadecenol and (E)-11-hexadecenyl-acetate) for monitoring the olive moth, *Prays oleae* and jasmine moth, *Palpita unionalis*, respectively. Sex pheromone catches (only males) were persistently lower in all collections of 2 successive olive seasons of 2002 & 2003 than those of UV-traps. *P. oleae* catches (males & females) in the UV-trap significantly ($P<0.01$) exceeded *P. unionalis* moths. The obtained results indicated that the UV-trap may represent an effective tool for mass-trapping of *P. oleae* and monitoring tool for *P. unionalis* and other species of moths in the area. The trap was so effective in trapping *P. oleae* even when adult population was low compared with sex pheromone trap. Some of non-target species may be protected from damage by placing a screen of proper sized mesh over the entrance of the trap. The UV-trap saved the fruit yield of a large olive farm without using any insecticides throughout the study period.

Classification of the Miridae (Hemiptera: Heteroptera): current status and need for future study

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The Miridae represent the largest family of Heteroptera, with more than 10,000 species. Despite their importance and the availability of a world catalog, the classification of this diverse family is far from stable. Currently, eight subfamilies are recognized: Bryocorinae, Cylapinae, Deraeocorinae, Isometopinae, Mirinae, Orthotylinae, Phylinae, and Psallopinae. The relationships among them, however, are uncertain and at least one of the larger subfamilies probably is not monophyletic. Phylogenetic relationships of the subfamilies and their tribes based on morphological attributes and trophic habits are discussed, and recommendations for further study are offered.

Adult development and behavior of Mediterranean fruit fly, *Ceratitis capitata* (Wiedemann) (Diptera: Tephritidae) recorded using digital photography at different constant temperatures.

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The development and behavior of adult Mediterranean fruit fly, *Ceratitis capitata* (Wiedemann) were examined using digital photography taken in growth chambers maintained at constant temperatures of 18, 22, 24, 26, 28, 30, 32 and 34 ±1°C. Fly maturity (days until calling, mating and ovipositing) was slightly shorter at higher temperatures (24 - 34°C) than at 22°C. However, the time to maturity was more than twice as long at 18°C than at 22°C. Calling by male flies occurred during the morning and early afternoon. Males stopped calling before 13h00 at temperatures below 24°C but the calling period was extended until after 14h00 as temperatures increased. These findings imply that temperatures below about 18°C are unsuitable for mass releases of sterile flies in SIT programmes. The frequency of mating and ovipositing increased during early morning and decreased again from mid-afternoon at most temperatures. Feeding on both protein and sugar took place throughout the day. Fecundity was highest at temperatures between 22 and 30°C. The number of days until 50% of the total number of eggs laid decreased from 34 days at 18°C to 8 days at 34°C. The minimum threshold temperature for oviposition was estimated at 12.22°C.

Biological control of pest thrips in perennial tree crops

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Pest thrips attack a variety of economically important tree crops in tropical, subtropical, and temperate climes. Typically thrips pest populations build rapidly, and larval and adult feeding damage to immature leaves and fruit and can be very serious resulting in significant economic losses. Many attempts have been to control exotic and native thrips pests with natural enemies. Control programs have included classical biological control programs against exotic thrips neoclassical programs against native pest thrips, and augmentative or inundative programs against native and exotic pest thrips with mass-reared natural enemies. To date there have been no successful or economically feasible control programs for thrips pests of outdoor tree crops. This presentation will examine the question "Why?" this lack of control is so consistent and prevalent. The avocado thrips, *Scirtothrips perseae*, avocado, *Franklinothrips orizabensis*, *Chrysoperla carnea* system in California will be critically examined to determine why biological control of this pest has consistently failed.

Attract and kill strategy to control *Carpophilus* spp. (Coleoptera: Nitidulidae): where are we now?

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We previously demonstrated that aggregation pheromone and co-attractant (fruit and fermented fruit juice) had considerable promise to control *Carpophilus* spp. by using attract and kill techniques. The trapping system used from 1999 to 2002 was messy and time consuming. To make the system more practical we identified the active components of the co-attractant and produced a synthetic version. Chemical analysis was conducted in the USDA – Peoria, IL laboratory and identified a series of aldehydes, alcohols and esters as potential attractant. We tested this with mainly *C. davidsoni* in the wind tunnel to understand how they respond to the smell from fresh and fermented fruit juice, and various blends of synthetic attractant. The best-performed blend of synthetic co-attractant was field tested to evaluate its performance. Synthetic co-attractant performed very well in the field and proved to be very species specific to *Carpophilus* spp. This facilitated easier and faster segregation and counting. Trapping efficacy usually depends on trap design as well as the chemical components (co-attractant). The results of an experiment to determine the best trap design to use for mass trapping of *Carpophilus* spp. will be discussed.

Understanding host-associated effects on oriental fruit moth, *Grapholita molesta* busck, biology: optimizing tree fruit IPM programs in the eastern united states.

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The Oriental fruit moth (OFM), *Grapholita molesta* Busck, has recently emerged as a major direct pest of apples in the eastern United States. OFM has traditionally been a pest of stone fruits, but since 1998, OFM infestations have resulted in over a thousand rejections of truckloads of apples from processing companies in the mid-Atlantic region. In order to better understand the reasons for these recent OFM outbreaks, a multi-faceted research program was conducted to explore the effects of host plants (apple and peach) on OFM survival, development, and behavior. Larval survival and development varied significantly between apple and peach. Additionally, host plants affected OFM oviposition behavior and possibly adult dispersal patterns between hosts in ways that could affect population dynamics at the orchard level. Also, degree-day based egg hatch models for both apple and peach were developed and validated based on daily egg hatch data collected in orchards. The results of our studies have allowed us to develop more host-specific models for OFM population dynamics and phenology and to tailor specific recommendations for OFM management on each host. This new information will allow growers to more effectively implement control tactics for the management of this pest on both hosts.

Integrated pest management in macadamias – opportunities and challenges

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IPM is a new approach working towards control of major pests in macadamia orchards in Australia. The two key insect pest of major concern for macadamia growers are currently the macadamia nutborer (*Cryptophlebia ombrodelta*) and fruitspotting bugs (*Amblypelta* spp.). Due to the problematic sustainability of repeated pesticide use in orchards, a pest management strategy with minimal input of chemicals is required. The proposed IPM strategy includes biological control, improved monitoring and IPM compatible pesticides. A current challenge is the integration of an egg parasitoid for control of *C. ombrodelta* into an overall IPM program. Improvements on monitoring tools for better indications on activity of fruitspotting bugs (*Amblypelta* spp.) female and *Amblypelta* spp. are required in the future for a successful IPM program. New developed insecticides for major pests in the orchard are being screened for their impact on beneficials to determine their compatibility with IPM. To ensure the practicality and adoptability of a new IPM program, involvement of crop consultants and growers is essential. Once the current key pests are being managed, adaptation to new emerging pests will have to be considered in the future. IPM needs to be regarded as a continuously adjusting process.

Integrated management of aphid on chilli (*Capsicum Annuum* L.) in Malaysia

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Chilli (*Capsicum annuum* L.) cultivation is a viable venture in Malaysia provided that the major pests and diseases on chilli can be successfully managed. The aphid, *Aphis gossypii* Glover has been reported as one of the major insect pests of chilli in Malaysia because it is a vector for CMV and chilli CVMV diseases. Majority of growers resort to use insecticide to control it that led to resistance problem. In Malaysia, studies on effect of crop variety and diversity, plant densities, mixed and inter-cropping, optimum chilli density, crop rotation and monitoring and use of reflective much on population dynamic of *A. gossypii*, its predators and virus diseases incidence have been conducted for years. Results were convincing and many of these IPM approach have been put into practiced by the growers and that incidence of virus diseases have been reduced. Interestingly, with the implementation of IPM in chilli cultivation, the cost of production was 11.8% lower than before IPM; with an annual chilli production about 4 000 ha, savings in the cost of production over US$ 2 million annually. Additionally, 13% increase in yield in chilli with IPM was reported that worth of US $ 1 – 2.5 million.

Development of sustainable area-wide mating disruption IPM in Victorian orchards, Australia.

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Mating disruption (MD) is a major tool of Integrated Pest Management (IPM) in Australian orchards. The successful use of MD to control oriental fruit moth (OFM) in Australian orchards started about 20 years ago and the approach was to treat with MD only individual orchard blocks and only known hosts. After that, some OFM damage at the edges of MD treated blocks stimulated the establishment of an area-wide MD approach. Such an area-wide MD program improved the protection of 40 contiguous orchards (1,100 ha) during two consecutive seasons in 1997-99. This program substantially reduced OFM numbers, prevented edge damage and helped many growers to reduce or eliminate insecticide sprays against OFM. Despite this success some growers did not continue MD applications after the project finished. OFM began to increase and this encouraged local growers to continue area-wide MD program as a self-sufficient community approach. They achieved re-establishment of MD across the whole region in 2002. To reduce the cost of the area-wide MD program, trials were conducted on the use of reduced rate of MD on pome fruit, barrier MD treatments, new MD products including sprayable microencapsulated and multi-species MD dispensers. Pheromone and semiochemical based IPM and selective area-wide MD programs are the key elements in development of sustainable strategies for multi-species pest control while protecting the environment by reducing pesticide pressure in orchards.

The importance of considering the impact of providing nectar sources on pests as well as natural enemies.

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The provision of nectar sources in the form of flowering plants has the potential to increase the level of biological control in orchards. Laboratory experiments concluded that the provision of flowers of buttercup, dandelion or honey solution increased the net reproductive rate of the leafroller egg parasitoid *Trichogrammatoidea bactrae fumata* when compared to provision of clover or water. In addition, the provision of honey solution increased the longevity and fecundity of a larval parasitoid, *Trigonospila brevifacies,* attacking leafrollers. Although field trials in an apple orchard revealed that the provision of *Alyssum* resulted in an increase in sticky trap catches of the leafroller larval parasitoid *Dolichogenidea tasmanica*, no corresponding increase in leafroller parasitism on the crop could be detected. The benefits of nectar sources increasing the reproductive rate of a parasitoid would be negated if the reproductive rate of the pest also increased. Subsequent trials showed that although the provision of honey solution increased longevity of the adult leafroller *Ctenopseustis obliquana*, it did not increase the pest's fecundity. We highlight the importance of the provision of plants that provide benefits to natural enemies but do not confer any benefits to the pest. We also emphasis the need to make the link to the desired end result - a reduction in pest damage.

Environmental friendly methods for controlling *Oryctes rhinoceros* **(Coleoptera: Scarabaeidae) in oil palm**

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The zero-burning replanting method is recommended for the oil palm industry in Malaysia. A slight drawback is that it provides an abundance of decomposing oil palm trunks which are very attractive for the breeding of rhinoceros beetles (Oryctes rhinoceros). To reduce this problem, monitoring and mass trapping should start a few months before replanting, in order to reduce the resident population and the immigration rate of the pest into the replanting area. The correct choice of replanting technique is very important. Underplanting is not recommended, as the standing dead trunks are most attractive to the beetles. The decomposing materials should be made less conducive for breeding, by spreading them in a single layer as mulch. In this manner, the decomposing material dries out quicker, rendering it unsuitable for breeding. Biological control agents in the form of Metarhizium wettable powder formulation have been developed for drenching in the trunk chips, giving effective control of the population up to eight months. A local virus strain which is most effective for O. rhinoceros can be released into the infested area by inoculation onto live adult beetles and re-releasing them. These biological control agents offer good alternatives to minimize use of chemicals.

Integrated pest management of western corn rootworm (*Diabrotica Virgifera Virgifera* Le Conte) in Bosnia And Herzegovina

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Western Corn Rootworm(WCR) is noted for first time in Europe 1992 and in B&H 1997. We start intensive farmer's education about WCR at the same time when WCR is noted in B&H for first time. In the beginning we mostly did lectures and last year we organised Farmer Field School (FFS) on cornfields already infected by WCR. All our activities made until 2002 were good base for FAO Project Integrated Pest Management (IPM) of Western Corn Rootworm in Central and Eastern Europe. B&H are also part of this Project as well as Hungary, Serbia and Montenegro, Romania, Bulgaria, Croatia and Slovakia. Project started officially 2003. We worked with 4 farmers groups. We educated them about WCR and ways how protect corn fields without pesticide use. Farmers came ordinary on the monitoring field, checked it, noted weather and soil conditions, WCR and other pests showing, damages on corn caused by different pests and at the end farmers discussed about results, made conclusions and decided on next meeting. Results, which we got during one year, showed farmers great interest for FFS education model and for IPM of WCR. These facts are very important for us in B&H where WCR is noted 1997 but there are not damages on corn caused by this pest.

Utilizing native biodiversity to enhance biological control

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Habitat management to enhance natural enemy efficacy has emerged as a central component of conservation biological control research and implementation. A survey of the recent literature on habitat management reveals a surprising reliance on plants not native to the region of study. This reliance on non-natives may be influenced by several factors including; availability, low cost, ease of establishment, and research indicating prior success in other environments. However, there is little ecological rationale to suggest that plants native to the region of interest cannot provide similar benefits. In addition, native plants may have unique ecological, economic and cultural values that favor their use. As science and societies move towards large-scale implementation of landscape management plans, the use of native plants in habitat management schemes takes on added significance. We describe research aimed at identifying native plants to provide pollen and nectar resources to insect predators and parasitoids and the development of strategic partnerships to facilitate their incorporation into agricultural landscapes.

Assessing insect dispersal with stable isotope techniques as a basis for accurate IPM decisions

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"Whether an IPM program should be implemented on a local (e.g. individual field) or regional basis (e.g. through the cooperative effort of several farm managers) depends in large part on the dispersal capabilities and behaviour of a focal pest and its key natural enemies. Because there is a relationship between the nitrogen stable isotope signature of a consumer and its diet, it is possible to infer whether a population has developed locally versus regionally by assessing the correlation between the nitrogen isotope signal of a field versus the nitrogen isotope signal of an insect population collected within that field. However, this analysis is complicated by the fact that immigrants quickly diverge from the signature of their natal field once they arrive and begin to feed in a novel field. Working in California cotton, we circumvent this issue by analyzing the wings and bodies of adult insects separately. Because the wings of adult insects are relatively inert, metabolically, contemporary feeding does not change the nitrogen isotope signature of the wings and they thus reflect natal feeding. Stable isotope analysis revealed that the herbivorous pest, *Lygus hesperus* (Miridae), exhibited local dynamics, as did predatory lacewings (Chrysopidae). Predatory damselbugs (Nabidae) exhibited regional dynamics, while big-eyed bugs (Miridae) and assassin bugs (Reduviidae) exhibited a mix of regional and local dynamics. These data suggest that *Lygus hesperus* should be managed on a local basis (e.g. field by field) and that three of its four key predators had foraged extensively within cotton fields where they were collected.
"

Maternal spread of *Enterobacter agglomerans* and *Klebsiella pneumoniae* through populations of the Mediterranean fruit fly, *Ceratitis capitata* (Weidemann): another piece of the symbiotic puzzle

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Enterobacter agglomerans and *Klebsiella pneumoniae* have been associated with fruit fly pests within four genera of Tephritidae. The associations include aspects of fruit fly nutritional, chemical, and behavioral ecology. The role these bacteria play in the life history of pest tephritids is not fully understood, however recent findings suggest that these bacteria are important in maintaining a healthy and functional gut. Probiotic diet formulations containing *E. agglomerans* and *K. pneumoniae* have been shown to improve the gut of irradiated male *C. capitata* and *A. ludens* used for SIT. In addition, sterile *C. capitata* have demonstrated improved mating efficiency after feeding on particular probiotic formulations. Reports of horizontal and now vertical transmission of *E. agglomerans* and *K. pneumoniae* strongly link these two bacteria to tephritid fitness.

Resource subsidies for enhancing the effectiveness of *Diadegma semiclausum* Helen, a parasitoid of the diamondback moth (*Plutella xylostella* (L.))

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When biodiversity is increased in agro-ecosystems, a range of benefits may follow. However, researching and selecting the most appropriate type of biodiversity is important. 'Resource subsidies' for predators and parasitoids can, for example, be valuable for the enhancement of natural enemy fitness, through aggregation on or near the added resource, changes in longevity, fecundity, parasitism rates and sex ratio and, ultimately and desirably, reduced pest populations. Many flower species have proven appropriate for this, with the ideal species depending on the agro-ecosystems. Selectivity of these can be achieved by using plant species that enhance a particular parasitoid's activity without enhancing the pest's fitness. The fact that many habitat manipulation studies have been done without considering possible positive effects on the pest population, and that plant selection has not often been rigorously tested, is an obvious limitation. In the current work, the effect of floral resource subsidies on the fitness of *D. semiclausum* and its host, the diamondback moth, was studied in the laboratory and field. Dispersal of the parasitoid from flower patches in the field was quantified, as was the parasitoid's effect on its host at a range of distances from the flower patches. The value of the appropriate selection of floral resource subsidies for pests' natural enemies and its relevance in IPM are discussed.

Successful IPM in protected cropping: cooperation between research and application

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Development of resistance shortly after pesticide use in greenhouses in the 1950s initiated research in Integrated Pest Management. The first natural enemies became available in the 1960s. A change from pure chemical control to very advanced IPM programmes could took place in the short time span of only 20 years, because of thorough basic and applied research, and intensive international collaboration. These IPM programmes were developed in close collaboration with all stakeholders, e.g. growers, biocontrol/IPM industry, extension service, plant breeders, entomologists, phytopathologists, micrometeorologists, simulation modelers and agronomists. The change to IPM was not based on idealism (cleaner or healthier environment), but was rooted on clear advantages for the grower. Nowadays, most if not all insect problems can be solved without use of insecticides in the main vegetable crops in northern Europe. Climate management, use of tolerant/resistant cultivars and recently biocontrol prevent/reduce disease problems. New activities in plant breeding aim at selecting plant cultivars that are able to attract natural enemies stronger after pest attack. Integrated management of pests and diseases without conventional chemical pesticides is a goal that will be realized in Europe within a decade (van Lenteren 2000 A greenhouse without pesticides. Crop Protection 19:375-384).

Key components involved in behaviorally based monitoring and management of flying and walking pests of tree fruit

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Integrated pest management approaches are becoming more prevalent in tree fruit production systems. These strategies include behaviorally based monitoring and control methods which rely on manipulation of a specific behavior of an insect pest in order to protect a valued resource. Identification of specific stimuli which result particular behaviors are the focal point for development of these strategies. A stimulus must be one that can be presented to the insect pest in a specific, perceivable, reliable, and practical form. In general, key components of behaviorally based monitoring and control methods include: (1) olfactory stimuli; (2) visual stimuli; (3) an effective capture mechanism; and (4) a decisive deployment strategy. However, these components differ widely among pests and particularly among pests which move differently toward host fruit trees, i.e., by walking or by flying. Comparisons of tree fruit pests belonging to Tephritidae and Curculionidae families will be used to highlight these fundamental differences.

The parasitism rate of leafrollers is influenced by the regional, rather than local, distribution of resources in New Zealand apple orchards.

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The availability of resources such as nectar or pollen can extend the longevity of parasitoids and increase the parasitism rate of prey populations. We provided the braconid parasitoid *Dolichogenidea tasmanica* with such resources in an attempt to increase the parasitism rate of the lightbrown apple moth larvae (*Epiphyas postvittana*) in apple orchards. Five replicated treatments of plots with different flowering plants were established within orchards. Parasitism rates of natural moth infestations and of those on sentinel plants were not significantly different from controls. However, parasitism rates were high, ranging between 51-94%. One hypothesis that may explain this result was that parasitoids freely dispersed between treatments within orchards. Further experiments examined how parasitism changes with distance from a large refuge population of parasitoids, and with a varying amount of local floral resources. Logistic regression analysis showed no significant effect of the availability of nectar, but a highly significant effect of distance from the refuge. Parasitism rates were on average 68% at 30m from the refuge, but declined linearly with distance to 21% at 300m. For this parasitoid-host relationship, effects of resources at the patch level appeared to be overwhelmed by a high regional dispersal ability of the parasitoid.

Management of oriental fruit moth with SPLAT-OFM A&K by ISCA technologies

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SPLAT-OFM A&K (ISCA Technologies, Inc. Riverside CA, U.S.A.), a proprietary biodegradable emulsified semiochemical dispenser containing *Grapholita molesta* (OFM) sex pheromone and small amounts of insecticide, proved to be economical, adaptable, easy to apply, and highly effective in controlling high OFM populations densities in commercial apple orchards. A single application of SPLAT-OFM A&K promoted continued trap shutdown, substantially decreased numbers of immature OFM findings, and improved the quality of fruit at harvest. Check plots under traditional management relying on insecticide applications sustained large populations of OFM, as reflected by large numbers of males captured in monitoring pheromone traps and larval damage.

Management of codling moth with ISCA Protec-Cydia by ISCA technologies

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ISCA Protec-Cydia (ISCA Technologies, Inc. Riverside CA, U.S.A.) is a low cost, low density, mating disruption formulation designed to effectively combat the quarantined pest *Cydia pomonella*, the Codling moth, present at medium and low densities in urban settings in Brazil. The proprietary formulation ISCA Protec-Cydia, consists of two components, a killing board and a sex pheromone based lure, to attract and kill males of *C. pomonella*. ISCA Protec-Cydia was applied on host trees of the quarantined urban zones at densities that varied between 12 and 60 units per hectare. Population data from pheromone-baited monitoring traps installed in the more than 7000 hectares under the mating disruption program indicate that the formulation ISCA Protec-Cydia effectively suppressed urban *C. pomonella* populations every time it was applied in the field, independent of the density of units per area. ISCA Protec-Cydia is an efficacious, safe and economical form of control of *C. pomonella* populations in urban settings.

"Montor system, an automated data collection and management system to effective support pest management decision making. "

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"The Moritor System (www.moritor.com, ISCA Technologies, Inc.) incorporates the latest innovations in information technology and engineering to give growers timely and accurate information about pests in their fields to support rapid, collaborative, pest management decision making. With ISCA's Moritor System, field data is collected with handheld or automated devices, sent to an internet accessible centralized database, where they are parsed, stored and analysed to generate reports which identify alarm situations and predict pest population densities, thereby facilitating decision making and the deployment of pest control measures in the field. Developed in part with funding from the National Science Foundation and National Institute of Standards and Technology, the Moritor System enables targeted, environmentally friendly pest management practices - the right pesticides at the right quantity, at the exact location, at the right time - specifically tailored to the user's needs. We will discuss the effect of Moritor's nearly real time field information on the increasing of precision in decision making and how it can foster interdisciplinary collaboration to micromanage insect populations on order to achieve effective low impact pest control.
"

SPLAT-OFM MD by ISCA technologies promotes long term mating disruption of *Grapholita molesta*, the oriental fruit moth (OFM)

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Although pheromone mating disruption offers notable advantages such as human and environmental safety, this tactic for managing moth pests in tree fruit crops has sometimes proven less effective than desired, precluding its use even under the current regulatory climate of shrinking insecticide availability. SPLAT (ISCA Technologies, Inc. Riverside CA, U.S.A.) brings a new brilliant solution to the pheromone mating disruption field, opening new perspectives to the formulation of economical and environmentally friendly, yet effective pest control solutions. SPLAT-OFM MD, a proprietary biodegradable emulsified semiochemical dispenser containing *Grapholita molesta* (OFM), proved to be economical, adaptable, easy to apply, and highly effective in controlling OFM populations in commercial apple orchards. A single application of SPLAT-OFM promoted over four months of nearly complete trap shutdown, virtualy eliminated OFM larval findings, and substantially improved the quality of fruit at harvest in the treated orchards. SPLAT-OFM is a good candidate to substitute traditional insecticides for the protection of orchards in more sustainable fruit production enterprises.

SPLAT-PNC MD, a paint ball formulation by ISCA technologies promotes effective mating disruption of *Acrobasis nuxvorella* (Lepidoptera Pyralidae) the pecan nut casebearer (PNC)

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"SPLAT-PNC MD (ISCA Technologies, Inc. Riverside CA, U.S.A.), a slow release matrix containing the sex pheromone of *Acrobasis nuxvorella*, the pecan nut casebearer (PNC) encapsulated in paint ball shells was applied to commercial pecan orchards to disrupt mating finding of this key pest. The use of CO_2-propelled paint ball guns to apply SPLAT-PNC MD allowed for the easy, fast and precise delivery of the mating disruption product to otherwise inaccessible points in the canopy of these often very tall pecan trees. Use of the Moritor Data Management System (www.moritor.com) allowed for fast field monitoring data collection and instantaneous automated internet reporting and distribution, thus allowing for efficient remote collaboration, project management and rapid, well-informed pest management decision making. The mating disruption treatment resulted in a substantial reduction in monitoring trap catch, and nearly eliminated PNC larval findings in the area, with no detectable damage to nuts at harvest. SPLAT-PNC MD is a prime candidate to replace the currently used chemical and biological insecticides in the management of PNC in commercial pecan orchards.
"

Mango flower thrips abundance and development and their impact on fruit set in the soconusco region of chiapas, Mexico

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The relationship between mango flower thrips and fruit set was assessed under three different environments in the Soconusco region of Chiapas, Mexico from November 1998 to May, 2000. Samples of mango panicles were taken weekly to identify as well as to assess abundance and development of thrips; number of thrips per date was transformed to thrips-days, an expression that involves both abundance and permanence of thrips on mango panicles. Fruit set was measured from the end of anthesis until harvest, and wild plants in orchards were sampled to determine presence of thrips. Mouthparts of immature thrips were observed by electron microscopy. Four *Frankliniella* species were found on both mango panicles (only while flowers were open) and wild plants; most of the thrips on mango panicles were immature individuals. No correlation was found between thrips-days and fruit set, therefore it was assumed that thrips do not influence yield. A round structure was observed at the tip of the mouthparts in immature thrips, which suggests they mostly feed on nectar. Because thrips were present on wild plants all year round, it was assumed that mango panicles function as a secondary host, providing a source of nectar and assuring survival of young individuals.

Control of pest thrips on economic importance crops and influence of wild plants for infesting and preserving natural enemies.

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Polyannual field trials and surveys have been carried on two economic importance crops, peach (in open field) and strawberry (as protected crop in tunnels), in two Southern Italian Regions (Basilicata and Calabria) to draft protocols of integrated control strategies for pest thrips. Three species of thripids have been studied: *Frankliniella occidentalis*, the most harmful on both crops, *Thrips meridionalis* and *Thrips major* only for peach orchards. Here, data referred to chemical action thresholds and the influence of wild host plants on infesting crops are reported and discussed. On peach, the relationships between the presence of larvae and adults of thrips in flowers and symptoms on fruits provide a mean threshold at 5% of infested flowers (1/2 thrips per flower corresponding to 25-30% of damaged fruits). Wild plants contribute to increase slightly the thrips populations on flowers and little fruits in spring but constitute important hosts of natural enemies (predators). On strawberry, 3-5 larvae or adults of WFT per flower can be considered as threshold value. Infestations of wild plants by thrips contribute to increase the WFT populations and do not provide any support for preserving natural enemies.

Lettuce IPM in Australia

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Integrated Pest Management is an approach that encourages continual improvement in pest management techniques. Although the concept has been in circulation for half a century, strong IPM systems have been developed in few vegetable commodities in Australia. This paper will discuss the approach taken in the past five years to develop lettuce IPM in Australia, with a primarily focus on management of insect pests. The range of tools and techniques will be discussed for the development of the IPM strategy and for adoption. Regional variation in approaches and uptake will be discussed. When the project begun the key pest in lettuce in Australia and the primary motivation for the project was *Helicoverpa armigera* but the introduction and spread of *Frankinella occidentalis* and *Bemisia tobcci* Biotype B, and the potential threat of *Nasonovia ribis-nigri* have become important drivers for lettuce IPM.

Development and field evaluation of a pupal-color based genetic sexing strain of the melon fly, *Bactrocera cucurbitae*

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The first genetic sexing system for the melon fly, *Bactrocera cucurbitae* (Coquillett), based on pupal color was developed. This system permits the separation of males (wild-type brown pupae) from females (mutant white pupae). Egg hatch averaged 42% for flies 2-5 weeks old, with higher rates (ca. 50%) for younger flies. Adult emergence rate was normal, averaging ca. 92%. Adult egg fecundity and fertility (both sexes) were very low (< 1% of normal) after irradiation at 100 Gy 1, 2, or 3 days before emergence. Adult flight ability from standard 20-cm tubes averaged ca. 65%. Irradiated females landed on, and oviposited into, zucchini fruit significantly less than non-irradiated females. Field cage survival tests indicated that the new strain survived as well (over 80%) as wild males over a 7-da period when provided with food and water. Mating tests indicated that males-only sterile flies mated significantly better than sterile males from bisexual (male and female) sterile populations. Males fed only sugar failed to mate at all with wild females, while males fed a low protein diet (6:1 sugar:protein), or fed the standard 3:1 diet, mated as well as wild males. Results of several successful SIT field evaluations in Hawaii in which induced egg sterility was monitored along with fly populations in traps, will be presented and discussed.

Options for managing thrips and tomato spotted wilt on flue-cured tobacco

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Several thrips management options were examined in replicated field trials to assess their impacts on the incidence of tomato spotted wilt virus (TSWV) symptomatic plants in flue-cured tobacco. Transplanting date influenced the incidence of TSWV, with a higher rate of infection in the earliest transplanted tobacco in most years. The insecticide Admire 2F and the plant activator Actigard 50WG applied as tray drench treatments to greenhouse produced transplants both reduced the incidence of TSWV. The combination of these two materials was more effective than either product alone. Foliar Actigard and Acephate treatments (at transplanting plus 3 or 4 applications soon after transplant) also reduced TSWV incidence. Thus, management options are available to suppress TSWV losses in flue-cured tobacco. Transplanting date (planting 2-3 weeks after the first tobacco is transplanted) plus tray drench treatments of selected materials are most effective in reducing TSWV; however, some early-season foliar sprays also can be effective in suppressing TSWV.

Evaluation of mating disruption in reducing the infestation of light brown apple moth, *Epiphyas postvittana* (Lepidoptera: Tortricidae), in citrus

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Light brown apple moth (LBAM), *Epiphyas postvittana* (Lepidoptera: Tortricidae), is a pest of economic importance of many fruit crops including apple, grape, and citrus in Australia. In citrus LBAM causes fruit drop and scarring and affects orange export to the US. Mating disruption is a proven technique in LBAM management in apple and grape. To assess the effectiveness of the technique in citrus, two field trials have been conducted during 2001-2003 in a 4-ha block of mixed Navel/Valencia oranges on an organic citrus farm in the Riverina, New South Wales, using the commercial mating disruption dispenser ISOMATE LBAM PLUS. A third repeat of the trial is in progress and will be finished by June 2004. In addition, mating disruption demonstration trials are being conducted concurrently on six citrus farms in the Riverina. Trials conducted at the organic site were monitored intensively with pheromone traps, sentinel females and infestation surveys. The results showed that, at the application rate of 364-728/ha, the mating disruption dispensers reduced pheromone trapping to almost undetectable levels for 4-6 months in the treated area. During this period, most sentinel females in the treated area failed to mate. More importantly, infestation by LBAM in the treated area was reduced by at least 50%. Similar results have been obtained from the demonstration trials.

Origins and recognition of pest thrips

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Pest control strategies presuppose that a particular pest can be recognised, and thus its migration into and population dynamics within a crop can be monitored. Bio-control strategies presuppose that the systematic relationships of a pest are clear, such that natural enemies of related species can be sought. With pest thrips these suppositions are often not valid. Many workers have difficulties in recognising thrips species, particularly when two or more are associated with a crop. Evaluating the contribution of different plant species to the maintenance of thrips populations is thus difficult. In a multi-species thrips assemblage host associations may differ between thrips species, also between years and seasons. Distinguishing between within-crop breeding and thrips immigration from outside a crop adds is another problem. Biological control programmes must consider, in addition to the population dynamics of these vagile pests, the multi-trophic problems involved between tospoviruses, plants, thrips and natural enemies. This talk will examine how our inadequate information on variation in biology and behaviour within and between species impacts on control systems, particularly those based on biological methods, and introduce a new recognition system for pest thrips species, based on a simple molecular method.

Sustainable integrated system for pecan and beef cattle and their effects on arthropod populations in flood-prone and non-flooded areas

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A three year study conducted in Oklahoma, USA assessed the effect of grazing legume or native companion crops on native pecan orchard floors in flood-prone and non-flooded areas of the state. Emphasis was on total system management of pecan, livestock and forage. The presence of legumes increased pecan and beef cattle gains; however, non-flood areas produced more pecan and more beef than flood-prone environments. During flood events, aphid populations were greater and beneficial organisms slightly lower in areas with native grasses compared to sites with legumes. Pecan weevil populations were higher in non-flood areas than in the flood plane. Numbers of weevils recovered in locations grazed by cattle were greater then in ungrazed sites. Adult populations of pecan nut casebearer were similar in flooded and non-flooded areas; however, these same populations were greater in grazed versus ungrazed sites. No differences were observed on larval populations of pecan nut casebearer across any treatments. The affects of companion crops on forage production and beef gain resulted in no supplemental nitrogen needs, particularly in flood-prone areas. Grazing, however, reduced nitrogen content for pecan production by 6-8%. These studies reinforce the benefits gained from growing a legume-based companion crop in pecan orchard systems.

Vain hopes and last resorts in field and forest pest suppression

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It is tempting to look back at the techniques that have been implemented for the suppression of pests of field crops and forest systems and wonder what practitioners were thinking about. Ancient approaches relied on superstitions and divine intervention. Approaches that rely on trapping, herding, burning, barriers, or nasty combinations of highly toxic materials have been used, rejected, and resurrected. Some of these ideas worked when there were no other alternatives. Others never worked at all, but memories are short and wishful thinking or desperation can be powerful forces when the sales pitch is promising. More recently, in part because of dissatisfaction with mixed results or negative impacts of high technology, there has been a return to the spiritual approach to pest suppression and we are left wondering how far we have really come.

IPM program development: role of the stakeholders

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The development and implementation of IPM programs in protected cropping is complex with a multiple of factors involved. It is no longer acceptable to operate in a vacuum, where research/extension personnel focus only on the management of one pest (e.g., western flower thrips). In addition, for an IPM program to have broad applicability across the various regions where a crop is grown, it should be evaluated in those regions. This suggests broad and comprehensive participation by growers in the validation and adoption of IPM programs. A new model for IPM development and implementation has been evolving over the past few years and focuses on the formation of teams of individuals who bring their own expertise to bear on the diversity that is IPM. In Canada for example, the Federal Government has recently organized their IPM research under a new national program team and provided new funding for development of reduced-risk pest control technologies and strategies. This has allowed for better partnering with the sector and linkage to other government and non-government organizations. IPM has also been identified as a Best Management Practice under the new National Environmental Farm Plan initiative, which is expected to increase support and adoption of IPM by producers.

Can Thrips be controlled with insecticides?

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Thrips continue to be major pests of numerous ornamental crops in the greenhouse and landscape as well as horticultural crops grown in the field. The impact of thrips has been exacerbated by 1) continued transmission of the tospoviruses by many species, and 2) increasing concern over the movement of 'new' pest thrips species around the world. The utility of traditional insecticides to effect thrips control in both the greenhouse and field will be reviewed. The development of insecticide resistance, compatibility of pesticides with IPM programs, and the influence of insecticides on virus control/spread will be discussed. With the world-wide amalgamation of agrochemical companies, registration of new pesticides has slowed dramatically. However, there has been renewed interest in the development/registration of 'biorational' products including pathogens and botanicals for thrips control and repellency. A roundup of these latest products, in addition to an objective evaluation of their potential for thrips control, will be presented. Finally, the potential role of utilizing materials that 'induce' plant resistance for control of thrips' damage and virus transmission will be discussed.

Management of cotton pink bollworm by mating disruption using Pb Rope®L

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IPM through mating disruption using sex pheromones offers a practical and ideal approach to combat cotton Pink bollworm, *Pectnophora gossypiella* (Saunders). Large scale field trials were conducted in India during 2001-02 and 2002-03 at farmers level to evaluate the suitability and efficacy of Pb RopeL (sex pheromone formulation of Pink bollworm manufactured by Shin-Etsu Chemical Co. Ltd., Japan). Single application at pinsquare stage of the cotton crop with 200 and 300 dispensers/ha of Pb-RopeL were compared with control plot . Pink bollworm moth catches, average rosette flowers (2-4%), Pink bollworm damage (10-13%), bad opened bolls (7-9%) were significantly less in Pb RopeL treated plots compared to those in the control plot. Seed cotton yield was higher in Pb RopeL treated plots (21 qt/ha) as compared to control plot (18 qt/ha). Pb RopeL plots recorded 35-40% higher net profit compared to control plot. Based on two season study, it can be concluded that Pb RopeL at 200 dispensers/ha applied once during pinsquare stage effectively checks Pink bollworm damage and increases quality seed cotton with higher net profit.

Integrated pest management in mango orchards using the green ant, *Oecophylla smaragdina*, (Hymenoptera: Formicidae) as a key element in the Northern Territory of Australia

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Mango is one of the most important commercial crops in northern Australia. Growers currently rely on toxic insecticides to control the insect pests, resulting in environmental and social problems. To reduce farmers' dependency on insecticides, a suitable IPM program is needed. Previous work showed that green ants can efficiently control the major mango insect pests, but they protect soft scales, damage fruits by their formic acid and disturb people picking fruits. Further research addressing these constraints showed that soft chemicals can reduce soft scale numbers without devastating green ants, the ant colony isolation reduces fruit damage by formic acid and water spray reduces the ant activity when picking fruit. A field experiment with two treatments (green ants plus soft chemicals (GS) and chemical insecticides (CI), each having three replicates) was conducted from 2001 to 2003 at Howard Springs in the Northern Territory. In two out of the three years, fruit quality was 20% higher in GS than in CI. Number of fruits or management costs /tree were statistically similar between GS and CI. During harvest, farmers had only minor problems with ant disturbance. An IPM model using green ants as a key element is discussed against 'organic' production.

Why and how to use weaver ants, *Oecophylla smaragdina*, (Hymenoptera: Formicidae) as a major component to manage insect pests in mango orchards in Northern Australia

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Mangoes are an important smallholder and commercial crop in Southeast Asia and northern Australia. To achieve high yield and quality, growers rely on regular use of insecticides to control the major insect pests, resulting in economical, environmental and social problems. Therefore, a better control program is needed. Weaver ants are known to control over 40 species of insect pests on many tropical tree crops, but they are not being used to combat mango insect pests, although they naturally occur abundantly in mango orchards. To determine whether these ants can be used as a major component of an IPM program in mango orchards, we have studied them in Australia, Vietnam and Thailand. Based on the results from field and laboratory experiments and long-term monitoring from 2001 to 2003 at six study sites in the Northern Territory, an IPM program of using weaver ants together with the use of soft chemicals and farming strategies has been established. This poster, together with a video display, will demonstrate which mango insect pest can or cannot be controlled by weaver ants, how to use the IPM program with the potential of producing 'organic' product, and how to stabilize weaver ant populations in mango orchards.

Habitat management for stemborer control in Africa: underpinning mechanisms and long term biotechnological opportunities

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To understand the mechanism by which lepidopteran stemborers are controlled by a "push-pull" strategy in maize production, chemical signals (semiochemicals), released into the air by trap crops and repellent intercrops, were captured from the air above the plants using entrainment into a porous polymer and analysed by gas chromatography coupled to electrophysiological preparations from the antennae of the pests and natural enemies, followed by behavioural studies. Attractant semiochemicals were identified following chemical analytical studies from trap crops such as Napier grass, *Pennisetum purpureum*, Sudan grass, *Sorghum vulgare sudanensis*, and also maize, and confirmed as such by behavioural studies assessing oviposition preferences. For the repellent intercrop molasses grass, *Melinis minutiflora*, additional semiochemicals denoting unsuitability as a host were identified, including (*E*)-ocimene and (*E*)-4,8-dimethyl-1,3,7-nonatriene. It was further discovered that increased parasitism of stemborer larvae in maize, with *M. minutiflora* as the intercrop, was caused by the nonatriene also acting as a recruitment cue for parasitoids, e.g. *Cotesia sesamiae*. These analytical studies, and the identification of the "push" and "pull" semiochemicals, are providing an important means by which to maintain quality of planting material and seed during expansion of the programme to new sites in eastern and southern Africa.

Characterizing a farming system in the central mountain region of Puerto Rico: IPM compatible?

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Farming systems research assists the successful generation and implementation of new IPM technologies. To develop effective IPM strategies there needs to be an awareness of the agro-ecological, cultural and socio-economic reality found within the target farm systems. There also needs to be an understanding of the parameters (i.e., the resources needed, the potential outputs and the inherent limitations) of the relevant IPM techniques under consideration. One effective way of summarizing this disparate information is the use of linear programming models. A socioeconomic study was conducted in the mountainous central region of Puerto Rico and, using the data generated, a linear programming (LP) model was developed. The LP model combined this information and gave a fair representation of the farming system. Most of the farm activity occurs during the Christmas period, while the summer months are normally a time of low activity. Poor, unstable markets seemed to be the biggest constraint to this system. Insecticide use is limited in many of the crops, especially roots and tubers. Any introduced IPM technique would have to focus, primarily, on keeping labor costs down. Protecting cabbage by using an IPM technique that included *Trichogramma* releases was not considered feasible by the model.

IPM of *Tanymecus dilaticollis* on maise in Romania

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The maize, cultivated on three million ha in Romania, is one of the most important crop, and the crop technology involves complex protection measures vs. pests and pathogens. The greatest damages are produced by *Tanymecus dilaticollis* attack even from the emergence. In order to avoid the damages or total crop compromising, the adoption of a complex measures system based on seed treating is necessary, as follows:
for areas where the preceding crop was a winter spike cereal, vetch-oats mixture, bean, soybean, pea or
linseed, the chemical treatments are not indicated, because the density registers low values till 3 individuals/m^2;
if the preceding crop was maize, sunflower, sugar beet or sorghum, the maize seed treating is
recommended with the following ingredient: carbofuran (8,750 g.a.i/t seed), thiamethoxam (3,150 g.a.i/t), imidachloprid (4,800 g.a.i/t), bensultap (12,500 g.a.i/t).

- in the case in which, after plant emergence, there are favourable climatic conditions for pest appearance, the application of one correction treatment during vegetation is recommended on seed areas treated with: thiamethoxam (25,0 g.a.i/

Insect behavioural ecology and the efficacy of habitat manipulation

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Insect population responses to diversified agroecosystems are highly variable, depending on the species of herbivores, natural enemies and vegetation involved. There is an urgent need to develop a mechanistic framework to understand and predict the response of herbivores and natural enemies to spatial arrangements of vegetation in agricultural systems. In this paper we investigate the sources of variation in insect response to spatial arrangements of vegetation in agroecosystems by using a combination of experimentation (with cabbage-associated species) and modelling. We introduce an individual-based spatially explicit simulation framework as a tool to investigate and predict insect spatial dynamics in heterogeneous environments. The simulation framework has a strong emphasis on state-dependent behaviour, cognitive and sensory abilities and movement of insects. Simulation results show that it is important to have a good understanding of the behavioural ecology of the target pest before one can determine whether the addition of highly attractive or repellent plants in a particular layout and density is likely to have an important effect on population density and hence potential crop damage. The reported wide variation in population response of herbivores to diverse agroecosystems is replicated in this study using different simulation scenarios by varying the behavioural features of the herbivore and ecological characteristics of the agro-ecosystem.

Pheromone mating disruption for codling moth management in apple orchards in the Elgin area of the Western Cape Province of South Africa

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Pheromone based mating disruption is widely used in codling moth (*Cydia pomonella*) management programs in apple orchards in the Elgin area, South Africa. Geographical plotting was used to analyse the spatial distribution of codling moth and its damage in orchards under mating disruption. Factors such as slope, wind and open spaces reduce the effectiveness of mating disruption. In the Elgin apple growing area all these factors are present. In addition there are usually three and sometimes four generations a year. Therefore, mating disruption can not be used as a stand alone control measure. However, since the introduction of mating disruption there has been a marked reduction in the number of codling moths caught in pheromone traps and in the number of sprays used for the control of this pest, despite high levels of resistance to insecticides. Fruit damage is recorded mainly in the vicinity of open spaces in orchards (homes, water points, etc), known infestation sources such as infested orchards and bin stacks, at top of steep slopes, and along the south east margin of orchards, or the margin intercepting the prevailing winds.

Influence of mineral oil and botanical extracts on Asiatic citrus psyllid, *Diaphorina citri* Kuwayama (Hemiptera: Psyllidae) in China

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Diaphorina citri is the vector of the devastating citrus disease *huanglongbing*. Biorational approaches to *D. citri* management are required both in its current range and in the event of incursion into Australia. Increasing the concentration of oil from 0.25-1.25% v/v applied to *Murraya paniculata* flushes (0.9 mL/flush) infested with adult psyllids in small enclosures increased mortality ($r^2 = 0.489$), with significant control achieved from 0.75%. Doubling oil spray volume lowered the concentration required to significantly reduce survival. A single application of 1% v/v oil spray to mature *Murraya* shrubs with heavy *D. citri* infestations resulted in a significant decrease in adult number. However, the application of sprays did not reduce the total psyllid population, indicating that at deposits applied, oil acted as a repellent and psyllids moved from oil sprayed to water sprayed control trees. When oil (0.5% v/v) and botanical extracts (1% v/v) were applied to citrus seedlings in a screened enclosure and adult psyllids subsequently released, the number of eggs laid on oil sprayed seedlings was significantly lower than on the control. The addition of *Ipomoea cairica* and *Wedelia trilobata* extracts to oil did not improve oil performance but the extracts alone did not provide significant control.

The problem of termite infestation in oil palm plantations on peat in Peninsular Malaysia

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Conversion of marginal soils such as peat for large-scale oil palm plantations in Malaysia is to meet industrial demand. However, peat is favourable for termite colonisation, thus infestation is of concern. This ongoing study aims to find better understanding of the pest behaviour, towards effective IPM in oil palm plantations in Peninsular Malaysia. Transects sampling and census in peat areas had retrieved termite species from several genera. Monitoring by using rubber wood stakes has detected various termite activities within plots of 500m^2 where colonies characterisations were possible upon detection of their unique excavation pattern. Census on palm stands found some assemblages utilising palm, indicating *Coptotermes curvignathus* as the major pest. Attacks originated from timber debris with visible mechanism of mudwork encapsulation around trunk that reflects some tropism. Infestation period on a stand varied, but shorter in immature palms. Detection of forager movement by staining technique identified clusters of infestation within a plantation block to have originated from a single colony. Infestation in a field was found to have a tendency of spreading towards fixed cardinal angles. Without disturbance, a colony will gradually infest palms that intercept their foraging tunnels, but changes towards physical anomalies, treatments and antagonists within the fields.

The Management of the tea mosquito, *Helopeltis antonii* infesting cashew in Sri Lanka.

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Cashew (*Anacardium occidentale* L.) is a crop considerable economic importance to arid and semi arid regions of Sri Lanka.. *Helopeltis antonii* seems to be found in areas where cashew is growing under conditions optimum for cashew production including the localities where intensive cultivation and the best management practices are followed. Data on the fluctuations in population density of the pests, collected from sample trees at weekly intervals, showed that its build up commences from October onwards synchronizing with the emergence of new flushes after the cessation of monsoon rains. The pest population reaches its peak during the blossom period in January. Highly significant negative correlations were observed between meteorological factors like rainfall, relative humidity, and minimum temperature and the population abundance of the pest, and positive correlations between pest population (adults and immature stages) and percentage of infested shoots and panicles. Endosulphan 35Ec was quite effective in controlling first instar nymphs as an exposure of ¾ hr completely killed the nymphs, but when the interval was 24 hrs a complete kill was recorded after 1 ½ hrs. Therefore it was found that application of Endosulphan as a high volume spray at the time of emergence of new shoots, panicles and fruit set was effective in controlling *H. antonii.*

The management of major insect pests *Bactocera cucurbitaceae* and *Aulacaphora spp.* in cucurbits under 3 intensive systems: integrated chemical and organic agriculture in southern Sri Lanka.

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Commercial vegetable production in Sri Lanka is intensive in forms of artificial inputs ex: high levels of fertilizer and pesticide use. However, in recent years there has been increasing emphasis on alternative methods ranging from low input systems to organic agriculture. The management of insect pests in commercial agriculture (Cucurbitaceae crops) was studied under 3 systems in Southern Sri Lanka: Integrated Pest Management, Organic and Chemical in Southern Sri Lanka in two cultivated seasons in 1998 and 1999. The use of neem-based products with predatory ants gave excellent control of fruit flies *Bactocera cucurbiteae* in organic agriculture system but it was not sufficiently active to manage *Aulacapora spp.* The effect of these 3 systems on management of the major insect pests is discussed. The incidence of major pests was significantly low in organic agriculture system in comparison to the other two systems used. The benefits and implications of the three systems on management of insect pests in commercial agriculture is discussed with emphasis on the management of insect pests in organic agriculture.

Non-target Hawaiian insects captured in Biolure® traps in native Ohea and Koa forests at different elevations.

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Control of economically important fruit flies (Diptera: Tephritidae) has been a major obstacle to expansion of Hawaiian agriculture. Efforts for area wide suppression of pest tephritid fruit flies through integrated pest management techniques, including utilization of the Biolure® fruit fly traps, have been on going across the Hawaiian island since 2001. As this program expands from local farmers fields to homeowner's backyards, the potential attraction of non-targets has increased due to closer proximity to native forests. The attraction of non-target Hawaiian insects to Biolure® fruit fly traps was assessed in a disturbed native Ohea and Koa forest across different elevations.

Insect behaviour and Bt crops

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Transgenic plants should not be used in isolation but as part of an IPM programme to reduce the risk of pest populations developing resistance to transgene products. Insect behaviour is a key factor to consider when integrating transgenic plants into IPM systems as it plays a crucial role in the effectiveness of natural enemies in controlling target and non-target pests in transgenic crops. Behaviour also affects which natural enemy species are likely to prosper and which may decline in abundance due to reduced host/prey availability. Resistance management strategies employed to delay the build-up of resistant pest populations not only require that the transgenic crop is highly effective in controlling the target pest but also that untransformed host plants (refuges) are planted in the vicinity to reduce the selection pressure and to allow mixing of susceptible with any surviving resistant pest genotypes. This paper will outline why behaviour of natural enemies is one of the key factors influencing if parasitoids and predators will have positive or negative effects on the evolution of pest resistance. Furthermore, results will be presented on whether mutations in pest insects that allow them to survive on transgenic plants affect their ability to escape natural enemy attack.

Fruit flies on the move: what we can learn from a landscape level perspective.

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Understanding the movement of insects into and out of commercial orchards is a critical factor in developing successful IPM programs. However, studying the movement of orchard pests between patches of suitable hosts requires an examination of individual behavior at the landscape level which can be difficult to obtain. Using the western cherry fruit fly, Rhagoletis indifferens (Diptera: Tephritidae), as a test insect, we modify an existing landscape dispersal model to a larger spatial scale and investigate the movement of flies between patches of cherry trees. Model results are then compared to data collected from a case study in which the history of the fly population spread is known. By determining the maximum inter-tree distance that cherry flies are likely to move, and the landscape structures that encourage or discourage movement, we can improve the quality of IPM programs for this pest.

Commercialization of insect behaviour modifiers: two case studies.

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Mating disruption has become a central pillar of integrated pest management programs for oriental fruit moth and codling moth in Australia as it is in west coast USA, Italy and South Africa. Significant fundamental research into mating disruption of both oriental fruit moth and codling moth was conducted in Australia. Similarly, the pioneering commercial development of mating disruption for both oriental fruit moth and codling moth occurred in Australia. Development of mating disruption products to the point of widespread commercial acceptance has depended on a number of factors. These include: (1) accurate identification of the sex pheromone structure, (2) availability of affordable active ingredients, (3) effective and user friendly controlled release systems, (4) selection of suitable pest targets for disruption, (5) species present in the orchard pest complex and (6) receptive end users. Comparisons are made of the population dynamics and behaviour of oriental fruit moth and codling moth as it affects their suitability as targets for mating disruption. Data on release rates of several kinds of dispensers and a critique for methods for measurement of pheromone release is presented. Pressures driving the acceptance of mating disruption and data on commercial use of mating disruption in Australia, west coast USA, Italy and South Africa is presented.

Efficacy of mushroom casing and compost treatment against the sciarid *Bradysia tritici* (coq) (Diptera: Sciaridae)

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The sciarid fly, *Bradysia tritici* is a pest of cultivated mushrooms and besides having pest status these flies are also reported to be associated with the spread of fungal diseases. The present study investigated the efficacy of insecticides and fungicides applied as compost and casing treatments. The results are discussed in relation to the combined effect of insecticides and fungicides to reduce the yield loses that could likely be caused by the infestation of *Bradysia tritici.*

Reduced risk IPM of thrips on greenhouse crops

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IPM of thrips on greenhouse crops (ie., vegetables and ornamentals) involves a multitude of approaches depending on the target pest, host crop, marketable product and the final targeted markets (domestic versus international). The ecology and population dynamics of thrips can vary considerably depending on the host crop, especially between ornamentals and vegetables. The complexity of plant structures and the abundance of pollen in ornamental crops often result in high pest reproductive capacity and difficulty in control measures actually contacting the different thrips stages. Some thrips species are the main vectors for plant pathogens, such as tospoviruses. In these situations, control strategies are usually either prophylactic (eg., screening) or fast acting (pesticides). Today, the emphasis in IPM of thrips on greenhouse crops is reduced risk control options. These options can be IPM-friendly pesticides, biological control agents, use of trap crops, cultural/sanitation practices, and physical exclusion/control. Monitoring programs and economic action thresholds are also becoming standard IPM practices.

Incorporating new/novel approaches into IPM in protected cropping

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Pest control strategies for greenhouse IPM are a constantly evolving and dynamic approach. Due to crop production under a relatively closed system and usually very sophisticated production practices, an integrated management approach is often very successful. Initially, control strategies were based on inundative releases of a few parasitoid and predatory mite species. Now control measures include mating disruption, trap crops, entomopathogens (fungi, bacteria and viruses), nematodes, sterile insect technique, and banker plant systems. Pest management is viewed as a tritrophic system with close interrelationships among pest, control agents and the host plant. The role of biotechnology in pest management is often controversial. The potential use of biotechnology in protected crop IPM will be discussed.

Pests of woody components of arid Agri-Silvi-Horti-Pastoral Production Systems: some issues in Indian context

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The production systems of the arid regions support large human and livestock populations in India. The arid agroecosystem has undergone major changes over last five decades, altering the pest spectrum of the production systems. The naturally occurring *Prosopis cineraria, Zizyphus* spp and *Tecomella undulata* have been the most common woody components of the arid production systems. Although these woody plants contribute immensely to the sustainability of the systems, there has been general ignorance about the injury inflicted by insect pests to them, the consideration for pest management being restricted to crop components alone. However, the dwindling density of some of the tree species has attracted attention of researchers and stakeholders. Insect pests and diseases have been attributed as major biotic factors responsible for the phenomenon. In recent years large-scale mortality of *Prosopis cineraria* has been witnessed in some pockets. Investigations implicated the injury of *Acanthophorus serraticornis* and related species for premature drying of trees in association with other factors. As *P. cineraria* serves as a virtual insurance for sustenance during the frequently occurring droughts in the arid regions, the losses have alerted the cultivators to protect this important resource. Wild jujube has gained increased importance in the wake of the technique of *in situ* budding of improved varieties of *Z. mauritiana* on the existing rootstock, transforming it into reasonably high value fruit crop. However, high incidence of pests on *Z. mauritiana* limits profitable cultivation. The wild jujube shares the common pests of the improved varieties, facilitating their survival and multiplication and hence the concern. Desert teak, *Tecomella undulata* is fast losing its population in the endemic areas with evidence of the contribution of the insect pests like *Stegmatophora* and *Patialus tecomella* interfering its development into tree. Restoration of the population of this species could be achieved only after addressing these problems. Against this backdrop there is an urgent need to consider developing packages to shield these valued resources from the menace of the pests and educate the ignorant peasant communities about the same. The article presents the status of the problem, its implications and the available solutions with possible future lines of action.

Both intraguild predation and alternative prey disrupt conservation biological control

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We have been examining the use of unplowed refuge strips to conserve predatory beetles and improve the biological control of root maggot (*Delia* spp.) flies on radish. The community of predatory beetles at our site includes several species of smaller carabid and staphylinid beetles (< 1 cm in length) that eat root maggot eggs, and one common larger carabid beetle (> 1.5 cm), *Pterostichus melanarius*, that eats smaller beetles but not fly eggs. We conducted a series of field experiments designed to address two questions: 1) does intraguild predation by *P. melanarius* disrupt conservation biological control, and 2) would schemes to selectively conserve smaller beetles be effective at improving fly egg control? In two initial experiments we found that densities of smaller beetles were lower, and densities of fly eggs were higher, in the presence of *P. melanarius*. However, attempts to improve fly egg biocontrol by selectively increasing densities of small beetles in the absence of *P. melanarius* were not consistently effective, because the suppression of fly eggs broke down if alternative prey (aphids) were present. Overall, these results suggest that both intraguild predation and the presence of alternative prey can disrupt conservation biocontrol in this system.

Arthropod pests of Australian olives

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With the rapid expansion of the Australian olive industry during the past decade, plantings have been established in many parts of Australia in both temperate and sub-tropical regions, and the majority of growers have little or no previous horticultural experience. A national project conducted over 2001-2004 aimed to identify the major olive arthropod pests in Australia through collection of field samples, a written grower survey and grower meetings, as well as monitoring of eight grove sites in five mainland states over a two year period. The project revealed that while Australia appears to be free from a number of cosmopolitan olive pests such as olive moth, *Prays oleae* and olive fly, *Bactrocera oleae* some species such as black scale, *Saissetia oleae* and olive bud mite, *Oxycenus maxwelli* are widely distributed. Australia also has several native pests of cultivated olive, including olive lace bug, *Froggattia olivinia* and Queensland fruit fly, *Bactrocera tryoni.* The paper will introduce the major Australian arthropod pests of olives, and will discuss options for their sustainable control.

Managing tomato russet mite, *Aculops lycopersici*, in greenhouse tomato crops

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Tomato russet mite is a common pest of greenhouse tomato crops in Australia during the summer months. Effective strategies to deal with this pest within an IPM program are needed. Some novel approaches are being undertaken in the area of reduced risk pesticides and use of indigenous predators such as phytoseiids and cecidomyiids.

Typhlodromips montdorensis (Schicha) (Acari: Phytoseiidae) - an effective predator for use against thrips in greenhouse vegetable crops

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The Australian predatory mite *Typhlodromips montdorensis* (Schicha) has been trialled in several crops of cucumbers, capsicums and tomato crops over several seasons. It is particularly effective against western flower thrips, *Frankliniella occidentalis* (Pergande), but also controls onion thrips, *Thrips tabaci* Lindeman. Rates of introduction are very low compared with the most commonly used predatory mite, *Neoseiulus cucumeris* (Oudemans). Two or three introductions of 10 per square metre are generally sufficient. This species operates best in warm, humid conditions.

Exploitation of male sex pheromone response polymorphism in *Helicoverpa armigera* IPM

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"Evaluation of responsiveness of *Helicoverpa armigera* males to blends containing 0-100 % Z-9-hexadecenal in Z-11-hexadecenal at various locations in India revealed that blends in the range of 3-15 % are significantly more attractive to males and the significantly most attractive blend(s) within this range change(s) with location and host. A hypothesis was tested that if pheromone polymorphism exists in *H. armigera* then catches in an area having traps with the usual 3% blend alone, would be significantly less than that of an area with traps baited with many blends. When equal number of traps either baited with 3% blend alone or with blends ranging from 2-15 % were deployed separately in two parts of a 170 ha institutional cotton field, the average trap catch/month was 49.8 males- (area with 3% blend) and 341 males- (area with 2-15 % blends). In another experiment covering 8 villages, even under farmer's field conditions use of multiple blends resulted in doubling of moth catches in cotton compared to the 3 % blend. Importance of this in IPM was emphasized; when on a farmer's field (6 ha), multiple blends based mass trapping eliminated thousands of males, effecting reduction in insecticide sprays used for pest suppression.
"

Thrips semiochemcals: a promising approach for thrips pest management

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The behavioural responses of walking and flying adult thrips (Thysanoptera: Thripidae) to chemical odours were studied to assess new options for management of these insects. Target thrips included the New Zealand flower thrips (*Thrips obscuratus*), onion thrips (*Thrips tabaci*), and Western flower thrips (*Frankliniella occidentalis*). Simple field bioassays, a Y-tube olfactometer and a wind tunnel were used to investigate fundamental aspects of thrips host finding behaviour (e.g. attractant vs arrestant responses), the physiological state (e.g. satiated vs starved) of the thrips, as well as differences in responses between walking and flying thrips. The spatial efficacy of chemical attractants was determined in field trials. The behavioural responses of thrips to approximately 30 chemicals (not previously known as thrips attractants/repellants) were tested with promising results. The use of these chemicals for improved thrips monitoring and trapping and for use with "push-pull" strategies in pest management and biosecurity is discussed.

Curious, cautionary, and inexplicable natural products used in pest control

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Interest in natural products is growing rapidly. Sales of dietary supplements alone (not even including all herbal remedies) increased from 3.3 billion dollars (US) in 1990 to over 9 billion dollars in 2001. Approximately one-half of the population in the United States uses these products. Given the size of the market, and an increasing interest in all things natural, it is not surprising that commercial products are being developed for a wide range of human and animal uses, including pest control. However, natural products are not always safer than synthetic products. Just because a plant has been used for centuries does not mean it is safe or even desirable. Plants contain many toxins, some of which we can use for pest control purposes. However, some of these toxins can cause significant human or animal health effects, and many deaths have been reported. A great many, if not most, of the compounds in plants used in pest control have not been scientifically evaluated for effectiveness and safety. This presentation discusses a variety of natural products used in pest control ranging from curious to cautionary to incomprehensible.

The landscape context of biological control

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Biological control has primarily relied on local facilitation of natural enemy populations, but recent studies found strong effects of landscape structure on the dynamic interactions between pests and their natural enemies. Here, we focus on case studies in two important arable crops, oilseed rape and winter wheat, showing how predators and parasitoids of major pests were enhanced by a high percentage of noncrop habitats in the landscape. Due to higher parasitism by wasps, pollen-beetle damage in oilseed rape was reduced in complex landscapes. In a two-year comparison, expansion or decrease in the area planted with oilseed rape led to a dilution or concentration of both damage by rape pollen beetles and its parasitism by wasps, respectively. Furthermore, parasitoids and predators may experience the surrounding landscape at different spatial scales. Parasitoids of cereal aphids were influenced at rather small scales up to 1km around a study plot, while highly dispersive linyphiid spiders responded to landscape structure in 2-3km surrounding. It is a major challenge for the future to understand the coaction of landscape effects on both pests and their multiple enemies, and thereby to identify applicable measures for landscape management that enhance natural pest control.

Challenges and strategies for controlling thrips spread of tospoviruses

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Increasing diversity among viruses in the genus *Tospovirus* (family *Bunyaviridae*) and their complex relationships with an increasing number of thrips vector species presents a global challenge to producers of ornamentals and food crops. Tospovirus epidemics require successful interactions between the thrips vector, the host plant for the thrips, and the tospovirus. Thrips vectors are usually highly resistant to chemical controls and there are no antiviral treatments to cure plants once they are infected. Therefore, successful control strategies must be based on interrupting the essential biological interactions. Given the global production of propagation materials, and international movement of cuttings and potted plants, pest managers must stay informed about emergence of new tospoviruses and new thrips vector species. The success of management strategies for thrips and the tospoviruses they transmit will depend upon a proactive approach aimed at prevention and a good understanding of the complex relationships occurring between the viruses and the thrips. This presentation will introduce the diversity of tospoviruses, the range of thrips species serving as vectors and highlight those aspects of their interactions that can be used in planning management strategies.

Effects of 'resource subsidies' on longevity of *Microctonus hyperodae***, a parasitoid of** *Listronotus bonariensis* **adults**

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Prior to the 1991 introduction of a biological control agent, *Microctonus hyperodae* (Loan) (Hymenoptera: Braconidae, Euphorinae), damage to New Zealand pasture by the Argentine stem weevil, *Listronotus bonariensis* (Kuschel) (Coleoptera: Curculionidae) was estimated to cost NZ$78 – 251 million per year. Potential exists to enhance the effectiveness of *M. hyperodae*, particularly in early summer when parasitoid adults fail to achieve much of their reproductive potential because they emerge up to four weeks before their hosts. It has been proposed that parasitism rates would be significantly increased if the longevity of these early-emerging parasitoid adults could be extended until *L. bonariensis* adults became abundant. Previous laboratory work has shown that the provision of sugar solutions can increase the longevity of *M. hyperodae* adults from 5 to 17 days. In this contribution, the effects of seven flowering plants on *M. hyperodae* longevity were measured in the laboratory. *M. hyperodae* lived a mean of 20.5 ± 3.5 days with buckwheat (*Fagopyrum esculentum* Moench), which was significantly higher than all other treatments ($P<0.001$). Survival analysis using a Kaplan-Meir survival function and Cox's Proportional Hazard Model for buckwheat compared with water showed 200% and 250% increases in parasitoid minimum and maximum survival, respectively. Results are discussed in relation to flower architecture and nectar quality, and future work is discussed.

Potential of behavioral components in insect pest management

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Abstract: In theory, Integrated Pest Management is the science and art of combining tactics into a strategy that will keep insect damage under economic thresholds. In practice, pesticides are, on a financial basis, the main tactics employed and this situation is unsustainable. Although deliberate use or manipulation of insect behavior is tactic that is seldom employed in IPM practice, recent advances in both science and technology should improve that situation in direct and indirect ways. The availability of new technologies, such as genetically modified cultivated plants, may directly effect changes in insect behavior of pests (and their natural enemies) such that the damage they would cause is under control. Indirect approaches concerns both pests and natural enemies. Knowledge of insect behavior per se and of the behavior of pests in relation to their nearby habitats can be used to achieve success in IPM programs. Transgenic natural enemies with improved fitness should be better adapted to perform in otherwise hostile agricultural environments. The future of IPM relies a blend of knowledge and technologies of which behavioral ecology plays a central role.

Ecology and feeding of the tarnished plant bug in vineyards

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Tarnished plant bug, *Lygus lineolaris* (P. de B.)(Hemiptera: Miridae), is a highly polyphagous insect of North America. Its presence in vineyards of Southern Quebec during all the growing season suggest that it may be a pest that warrant insecticide treatments. We first studied the migratory activities of adults with sticky traps deployed all around and in the center of a vineyard. Captures were recorded weekly. Secondly, to assess potential damage by tarnished plant bug during bloom, we videofilmed the feeding behavior of nymphs and adults on inflorescences. Localized feeding wounds were fixed and prepared for electronic and scanning microscopy. Histological studies concluded that during bloom time, feeding is restricted to the nectaries, which do not have functional relationship with ovaries. Thridly, cage studies done on part of plants covered with fine muslin revealed that tarnish plant bug decrease damage at stages 15 and 31 (Eichhorn-Lorenz phenological scale). Overall the impact of tarnished plant bug depends on the phenology of the plant.

Using HPLC to assess nutritional state, sugar sources consumed, and feeding history of field collected insects.

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Adult parasitoids depend on sugar-rich foods like nectar and honeydew to meet their energy requirements. The potential fitness benefits of sugar feeding for parasitoids have been established in numerous laboratory studies. Nevertheless, we know little about the nutritional ecology of parasitoids in the field, chiefly due to the limited specificity of methods applicable for field studies. Here we use HPLC sugar analysis to study the sugar profile of *Cotesia glomerata* and *Microplitis mediator* relative to feeding-treatment. We identify the overall sugar level and the ratio of glucose to fructose as two parameters that in combination unambiguously characterize an individual's nutritional state and feeding history. In addition to revealing past feeding events, HPLC analysis also identifies and quantifies a wide spectrum of sugars present in individual specimen. When food sources contain specific sugars, so-called 'signature sugars', this can provide additional information about the type of sugar source consumed. In our field study, we collected *Cotesia glomerata* and *Microplitis mediator* in a cabbage field with flowering herbs in the field margin, as well as in a control field bordered by grasses. HPLC analysis of the collected individuals revealed that >The sugar reserves of parasitoids collected in proximity of the flowering herbs exceeded the reserves of parasitoids collected in the control field. >80% of *C. glomerata* and 55% of *M. mediator* had consumed honeydew

Artificial food supplements for improved IPM: New results and a review of the evidence

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Artificial food supplements are a habitat manipulation tactic used in integrated pest management (IPM) systems to increase predator and parasitoid abundance and rates of predation and parasitism. The efficacy of using a single late season application and nine season-long applications of the commercial artificial food supplement Amino-Feed UV in cotton was assessed in two field trials. Measurements were made of entomophagous and pest arthropod abundance, crop yield and fibre quality in both trials, and mortality rates of sentinel prey and fruit counts in the second trial. In both trials, none of the above parameters was significantly affected by Amino-Feed treatment. To place these results in context, a hierarchical approach was used to review 82 trials from 38 publications on artificial food supplements. The hierarchical levels most commonly examined were the abundance of entomophagous (n = 51 trials) and pest (n = 26) arthropods. Although the abundance of entomophagous arthropods increased in 34 trials and the abundance of pest arthropods declined in 14 trials, increased profitability was not demonstrated in the four trials where it was examined. The likelihood of a positive outcome in a trial was not significantly affected by annual or perennial cropping systems, single or multiple applications of the supplement, or carbohydrate and/or protein components of the supplement. Recommendations for the direction of future artificial food supplement research are outlined.

The M3 bait station for fruit fly control – the IPM solution.

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A bait station has been developed for the control of Mediterranean fruit fly (*Ceratitis capitata*), Natal fruit fly (*Ceratitis rosa*) and marula fruit fly (*Ceratitis cosyra*). Effectiveness of the control measure was demonstrated using both male and female lures placed outside, on the perimeter and in the centre of the orchards. The outside traps indicated the fruit fly "pressure" while those in the orchard indicated the effectiveness of the control measure. Fruit inspections confirmed the effectiveness of the treatment. The M3 bait station IPM compatibility rests in the facts that it does not attract many non-target insects (no repercussions), food and environmental safety is ensured as no pesticides are applied directly to the fruit, tree or the environment. Other plus aspects of the system are that only a single treatment is required in a season, it is not affected by rain, it is easy to use and requires no skilled labour or sophisticated machinery, it is an alternative to organophosphate treatments, it is EUREPGAP compliant and finally it is cost effective.

The influence of various intercropping ratios of cabbage and carrot plants on the incidence of important cabbage insect pests.

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Integrated Pest Management (IPM) employs various strategies and relies on sound management for successful implementation. Shortages of quantitative information inhibit IPM efficiency. A trial was started at Potchefstroom (South Africa) during the 2001 / 02 season to address the problem. The aim, to determine the effect various intercropping ratios, involving cabbage and carrot plants, has on diamond back moth (*Plutella xylostella*) infestation and parasitism levels, as well as on aphid infestations. The trial consisted of five treatments *viz.* control (cabbage monoculture + chemical control), monoculture (cabbage), inter crop 1:1 (cabbage: carrots), inter crop 1:3 and inter crop 1:5, replicated six times. Insects were sampled weekly after removing leaves of similar age from plants. *P. xylostella* larvae were lab reared at temperatures above 20°C to determine parasitism levels. Aphid numbers were expressed per leaf area to allow for comparison between treatments. Feeding damage to cabbage plants was determined by calculating the R – Index of Attack for data from visual ratings. Preliminary results indicate no significant ($P \leq 0,05$) differences between *P. xylostella* infestations on monoculture and intercropping treatments. A high correlation ($r = 0,828$) existed between parasitism and *P. xylostella* infestations. No significant feeding damage differences ($P \leq 0,05$) were recorded between monoculture and intercropping treatments.

Spatio-temporal distributions of pests and their key parasitoids in a crop of winter oilseed rape: implications for integrated pest management

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The spatio-temporal distributions of four major pests (*Psylliodes chrysocephala, Meligethes aeneus,Ceutorhynchus pallidactylus* and *Dasineura brassicae*) and of their key larval endoparasitoids were studied in a crop of winter oilseed rape. Insects were sampled at 40 spatially-referenced points within the rape crop and the emergence of those that overwintered in the field was monitored in the following crop of winter wheat. Spatial distributions were mapped, analysed and compared using Spatial Analysis by Distance IndicEs (SADIE). The distributions of most pests and parastoids were complex with differing irregular patterns of aggregation. Some pests were markedly more edge-distributed than others. The environmental factors and behavioural responses of both pests and parasitoids that influence their spatial and temporal ecology will be discussed. The potential value of spatio-temporal information for the development of integrated pest management strategies designed to conserve parasitoids for bio-control by spatial and temporal targeting of insecticides will be considered.

Exploitation of herbivore-induced plant volatiles for conservation biological control of *Lygus* species

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Herbivory induces production and release of plant volatiles that attract natural enemies of the herbivores. Using plant volatiles to manipulate behavior of beneficial insects may be a powerful tool to enhance conservation biological control of crop pests. To provide a better understanding of this phenomenon and its potential for pest control, we have conducted a series of studies with *Lygus hesperus* and an egg parasitoid, *Anaphes iole*. Our ultimate goal is to apply knowledge of plant volatile production toward biological control of *Lygus*. Initial studies using GC-MS demonstrated that plants damaged by *Lygus* feeding and oviposition emitted a complex mixture of volatile organic compounds that is released systemically throughout the plant. These results led to electroantennography studies that showed that *A. iole* perceived many of the plant volatiles released after *Lygus* damage. Heightened antennal sensitivity by female wasps to several induced volatiles suggests a role in host habitat location. Subsequent behavioral studies (olfactometer and flight cage) in the lab and greenhouse demonstrated that female *A. iole* were attracted to plants damaged by *Lygus* and to synthetic components of the plant volatile blend. Studies are currently underway to determine if synthetic components of *Lygus*-induced plant volatiles are attractive to *A. iole* under field conditions.

Responses of cotton to actual and simulated green mirid damage.

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Green mirids damage squares (flower buds) and bolls (fruits) of cotton. They are an increasing problem during fruit set in crops with reduced insecticide applications, such as transgenic cotton, but their effect on profitability is poorly understood. We investigated compensation for mirid damage and the effects of mirids on yield, maturity and fibre quality. Crops with high mirid numbers (up to 6/m) over 50 days during boll filling had yields half those of crops with low mirid numbers (up to 1.4/m). There was no evidence of intra-boll compensation. We injected pectinase, a component of mirid saliva, into 10-day-old bolls, mimicking mirid feeding. The seed cotton mass of injected bolls was reduced by 38%. Artificially damaged plants compensated by increasing the size of undamaged bolls indicating that compensation can be achieved through the re-allocation of resources from damaged to healthy bolls. Injection of pectinase into older squares induced parrot beak boll formation, similar to that caused by mirids. The proportion of parrot beaks was 8 – 34% of the injected squares and varied between varieties. There was no evidence of reduced fibre quality because ginning appeared to effectively remove damaged lint so the risk of a penalty for poor quality was minor.

Habitat manipulation for IPM

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The efficacy of biological control as an ecosystem service can be improved by the provision of pollen, nectar, alternative prey/hosts and/or shelter. When flowers are provided, there is potential hierarchy of behavioural and population consequences. The hierarchy is: 1. Parasitoids aggregate on the flowers. 2. Parasitoid fitness (e.g., longevity, fecundity, searching efficiency) is enhanced. 3. Parasitism rate is increased. 4. Pest populations are decreased. 5. Pest populations are brought below an economic threshold. Achieving all five steps in the above hierarchy is difficult – this is compounded by the need for selective floral resources which ideally benefit the parasitoid but not the pest. However, recent research has reveled this “selectivity” also to have a hierarchical component. 1. Parasitoid benefits but pest does not. 2. Parasitoid and pest benefit, but the pest has higher longevity only. 3. Parasitoid and pest benefit but parasitoid benefits relatively more. 4. Parasitoid and pest benefit but the improved fitness of the pest leads to a further improvement in the parasitoid’s fitness because the developing parasitoid larva derives a higher level of nutrients from a host which has fed on, e.g., pollen. The last of these possibilities represents the novel concept of “indirect resource subsidies”. Examples of recent research illustrating the above two hierarchies in conservation biological control will be presented.

IPM in organic systems: an industry/university collaboration

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Ecosystem services (ES) are those services provided by living things for the good of mankind. They include such processes as pollination, biological control, soil fertility maintenance, carbon dioxide sequestration etc. All agriculture depends on ES but in organic systems, these services assume a greater importance. "Western agriculture" has consistently reduced the provision of ES with its extreme reduction in biodiversity. In July 1999 a commercial organic cropping farm was established at Lincoln University, New Zealand as a commercial, research, demonstration and teaching unit for the university and for the company. Also, it has been the focus of a sustained programme of enhancing ES on farmland, with an emphasis on ES provided by insects. Pollinator populations have been enhanced and the efficacy of arthropod predators and parasitoids has been increased. A "Biodiversity Trail" has been established on the farm to demonstrate these enhanced ecosystem services.

Does pest management in organic agriculture work differently? Case studies in European organic research

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Organic agriculture was founded by Rudolf Steiner and Hans Mueller in the years between 1924 and 1940, but it was not until the 1980s when pest management researchers began to develop concepts to control pests in organic systems. Today, insect pest management in organic agriculture involves the adoption of scientifically-based and ecologically sound strategies as specified by international and national organic production standards. These include a ban of synthetic insecticides and genetically modified organisms (GMOs). The first phase of an insect pest management program for organic systems is the adoption of cultural practices including diverse crop rotation, enhancement of soil quality by incorporation of specific cover crops and/or the addition of soil amendments, and choice of resistant varieties that help to prevent pest outbreaks. In the second phase, habitat management (e.g., incorporation of hedgerows and wild flower strips) is implemented to encourage populations of pest antagonists. Third and fourth phases of the program include deployment of direct measures such as biocontrol agents and approved insecticides. However, the strategies for pest prevention implemented in the first two phases often obviate the need for direct control measures. The researcher and grower approaches to pest management in organic systems are different than in conventional agriculture because, conceptually, the indirect or preventative measures form the foundation of the system and direct, or re-active control methods are rare and must comply with organic production standards. The range of European research activities for these concepts is illustrated with case studies in organic apple, corn, and vegetable production.

Prediction of biological control of the greenhouse whitefly by combined use of *Encarsia formosa* and *Eretmocerus eremicus*

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Imported parasitoid species, *Encarsia formosa* and *Eretmocerus eremicus* have been commercialized recently in Japan for controlling whiteflies on tomato crops in greenhouses. The combined release of these two parasitoid species might be applied as more effective method than the single species release of *E. formosa* or *E. eremicus*. To evaluate the effectiveness of this combined use to control the greenhouse whitefly on tomatoes, the response of *E. eremicus* on the whitefly and the interaction between *E. formosa* and *E. eremicus* were studied in laboratory or in glasshouse experiments. *E. eremicus* attacks all the whitefly instar larvae except the young crawling stage and the late fourth instar stage. The functional response to the whitefly larvae infesting tomato seedlings was studied using cages in glasshouse experiments. The functional response was Holling's type 2 response for all host larval stages. When whitefly larvae were parasitized by both *E. formosa* and *E. eremicus*, *E. eremicus* always survived and *E. formosa* was killed in the direct interference between two species. A simulation model was developed to predict the biological control when both parasitoids are released to control the greenhouse whitefly on tomatoes. When both species are released simultaneously of different release ratios, the intermediate ratios result in better control than the single species release of one of the two species.

Moth control using yellow fluorescent lamps

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The yellow fluorescent lamp (YFL) is a main physical control implement for integrated pest management (IPM) in Japan. YFL is used for overnight lighting. When the nocturnal moth adults enter the lighting area, they undergo "light adaptation", thereby becoming inactive. Radiation specifications of YFL are a wavelength range of 500–700 nm with a peak of about 580 nm. Such light does not attract most insects, yet it retains the potential for inducing "light adaptation". YFLs have been used in orchards against the fruit-piercing moths *Adris tyrannus* and *Oraesia excavata* or the like since the 1970s. Over the last decade, this implement has become particularly useful for vegetable and flower producers in order to control nocturnal moth oviposition. Moth species showing marked YFL effects are *Helicoverpa armigera*, *Spodoptera exigua*, *S. litura*, and *Hellulla undalis* etc. Crops of practical use are tomato, strawberry, cabbage, eggplant, carnation, rose, etc. Compared with a no-lighting condition in tomato practice, YFL reduced damaged fruits by 90% using only 20% of usual insecticides. Illuminance on the crops should be maintained at over 1–2 lx for the control effect.

Industry perspectives on research priorities for IPM in organic crops

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Crop production and pest control methods in organic agriculture are governed by strict standards and rules imposed by IFOAM or the USDA. Enhancement of soil quality and crop rotation form the foundation for organic agriculture and are effective strategies for controlling pests in annual crops. However, these methods are not always successful in controlling pests. Where they are not helpful, other tools, such as approved pesticides are needed. It is unlikely that organic agriculture will have a wide range of accepted chemical controls similar to what we currently use in conventional agriculture; therefore, the role of insect ecologists and behaviorists will become increasingly important in the development of new methods of crop protection. It will likely fall to them to develop or refine better scouting methods and thresholds. They will also need to improve our understanding of the crop ecosystem both above and below the soil surface. Finally, it will be their task to reevaluate our current agricultural cropping systems, designing new systems for organic agriculture where the crop environment discourages pests or allows predators to proliferate.

Introduction: IPM in organic systems

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IPM principles, strategies and tactics are highly applicable and adaptable to organic agricultural systems. Conventional farmers and ranchers are increasingly interested in approaches to IPM for organic systems since the release of USDA organic certification standards in 2002. Current applied research and extension programs are providing successful field-based working models of pest management for organic growers. Symposium speakers will give an overview of successful IPM programs that have been developed for organic field crop, fruit, and vegetable production systems. Research and educational needs will be identified to support the growing organic agriculture field for small and conventional-sized producers.

Effects of intraspecific competition on the levels of parasitism and progeny production between parasitic wasps for *Callosobruchus chinensis* host larvae

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Effects of intraspecific competition between two solitary parasitoids, *Anisopteromalus calandrae* (Howard) and *Dinarmus basalis* (Rond.) have been assessed in respect of parasitism and progeny production. The parasitoid, *D. basalis* demonstrated significant increase in parasiutism and progeny production over *A.calandrae* while confined to 3rd-4th instar host larvae *of Callosobruchus chinensis* (L.) in different durations. The individual mated female *of D.basalis* produced 9.1 ± 3.66, 13.9 ± 3.38, 20.8 ± 2.44, 23.1 ± 9.98 and 31.9 ± 19.58 progeny during 24,48,72,96 and 120 hours duration, respectively compared to 7.8 ± 1.99, 6.8 ± 3.85, 10.3 ± 6.56, 15.5 ± 7.26 and 24.8 ±13.05 progeny produced from *A. calandrae* in the above mentioned durations. The highwest parasitism of 50 % and the lowest parasitism of 4 % in 24 and 120 hours duration, respectively were observed *in D. basalis*. A maximum sex ratio of 2.49 and 2.75 (female to male) in *A.* calandrae *and D. basalis*, respectively were also noted.

The effect of wheats differing in category of resistance on Russian wheat aphid biology

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Biology of Russian Wheat Aphid (RWA) *Diuraphis noxia* (Mordvilko) was investigated on three wheat genotypes varying in aphid resistant. The aphid density was low on resistant than the susceptible wheat genotypes. Wheat Genomic differences affected the RWA developments. Antibiosis was observed as primary resistance category for wheat variety tested. A very high correlation was found between the number of aphids and the levels of infestation. However, the initial high level of infestation significantly affected the plants and aphid parameters as compared to the low level of infestation. Also more parasitoid were observed on TAM 107 than on resistant wheat genotypes. High level of RWA infestation affected the quality of seeds significantly in CO940626 and TAM 107. However, increased protein level was observed in susceptible wheat which might be due to aphid stress or lower test weight.

Biological control of peelminer in California citrus

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Peelminer, *Marmara gulosa* Guillen Davis (Lepidoptera: Gracillariidae), has been a persistent pest of citrus fruit in the San Joaquin Valley in central California since 1999. Larval stages create serpentine mines scarring the rind of citrus fruit rendering it unacceptable for fresh market sale. Erratic population increases and unexplained population movement into citrus and 65 other plant species from 28 different families leave many questions to be answered about this insect. Equatorial fruit diameter studies on grapefruit, pummelo and navel oranges showed no relationship between diameter and timing of egg deposition or mining initiation. Mining on oleander stems, *Nerium* spp., was recorded weekly from August to December of 2003 and showed significant population increases into the winter. Parasitoids of peelminer native to the San Joaquin Valley of California were composed only of eulophid species and have not demonstrated any population control. Peelminer also has been a persistent pest of citrus in the Coachella Valley of southern California since 1995. A gregarious eulophid parasitoid, *Cirrospilus coachellae* Gates, native to Coachella was released into the San Joaquin Valley starting in 2001. Successful establishment has not yet been observed in the San Joaquin Valley following the annual releases.

Effect of harvest management on pest and beneficial insects in alfalfa

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The alfalfa weevil, *Hypera postica* (Gyllenhal), and potato leafhopper, *Empoasca fabae* (Harris), are important pest of alfalfa, *Medicago sativa* L., in Missouri. Many producers trying to avoid insecticide use and conserve beneficial insect populations have attempted to reduce pest insect numbers by early harvest of the crop. Two of the most common harvest strategies employed for insect control are mechanical harvest and forage removal by grazing with livestock, usually cattle. In this study, pest and beneficial insect populations were estimated prior to and after each of four harvests of alfalfa annually. Treatments included all harvests by mechanical means, all harvests by grazing with cattle, and two treatments with mixed harvest methods regimes. Data indicate that larval numbers of alfalfa weevil were reduced by approximately 99% for the all-mechanical or all-graze method of harvest. Potato leafhopper adults were reduced by 86% by mechanical harvest as compared to 67% in grazed plots. Similarly, numbers of potato leafhopper nymphs were reduced by 83% and 18% for mechanical and grazed harvest, respectively. A complex of 10 beneficial insects were grouped for statistical analysis. Insects in this complex were reduced by an average of 81% in plots harvested mechanically as compared to 64% in grazed plots.

Seasonal fluctuation of adult and larvae of *Agriotes spp.* **(Coleoptera:Elateriadae) in central Croatia**

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The seasonal larval and adults abudance of the five wireworm species was examined in central Croatia in a three-year period. Sex pheromone and soil bait traps were used and soil samplings were made. Sex pheromone traps operated from April to September. Soil bait traps were installed for 15 days in April. Soil samplings were made in April at a depth of 30 cm. The population density of *Agrotes lineatus* is described as high at both localities. The swarming period of *A.lineatus* lasted from April to mid August. The population density of *Agriotes ustulatus* on two lacalities was from low to high. The swarming period of *A.ustulatus* lasted from mid May to early July. The population density of the *Agriotes brevis* at both localities is described as medium. Before this investigations there was no literature data about A.brevis presence in Croatia The population densities of *Agriotes sputator* and *Agriotes obscurus* were low to medium. The swarming period of all three species lasted from April to July. In soil samples 1-12 larvae/50 soil samples were found. In bait traps 2-111 larvae/50 traps were found. Using soil bait traps and soil sampling larvae of all five species were collected.

Implementation of an environmentally friendly IPM program for the control of the most destructive pest of coffee crops: the coffee berry borer *Hypothenemus hampei.*

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An enormous research program aimed at controlling *Hypothenemus hampei* (Coleoptera: Scolytidae), the most destructive insect pest of coffee, has been initiated at the National Coffee Research Center - CENICAFE, the research branch of the National Coffee Growers Federation of Colombia. After the introduction of this insect in Colombia in 1988, CENICAFE started an ambitious program to implement an environmentally friendly IPM program focusing on cultural and biological control. Since then, cultural control strategies have been successfully implemented. Moreover, three African parasitoids have been imported, reared, and tested in the field. Research on entomopathogens has involved the use of *Beauveria bassiana, Metarhizium anisopliae,* and *Paecilomyces lilacinus*. Field and lab experiments have yielded impressive results, including the development of a genetically modified strain of *B. bassiana* with *M. anisopliae* genes that increased virulence and shortened mortality time. Other experiments have consisted of the estimation of the worldwide genetic variability and biogeography of this insect and the detection of *Wolbachia* as a probable distorter of gene determination. Ongoing research focuses on exploring the use of pathogenic nematodes. Studies on butterfly biodiversity in coffee habitats and the morphological description of the alimentary canal and the reproductive system of *H. hampei* females will be presented.

Evaluation of different plants as an ovipositional substrate for *Orius insidiosus* (Hem : Anthocoridae).

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Orius insidiosus (Say) lays eggs inside and inserts them in the plant tissue. An obstacle for its massal rearing is the selection of a plant that is suitable as an ovipositional substrate, being one less expensive and easy to get and well accept by the female. The objective this work was to evaluate the preference and the suitability of different structures of several plants as ovipositional substrate for _O. insidiosus_. The trials were conducted in climatic chamber at 25±1ºC, RH 70±10% and photophase 12h. The plants site evaluated were amaranth stem (_Amaranthus virides_ L.), common bean's stem and pod (_Phaseolus vulgaris_ L.), green bean's pod (_Phaseolus vulgaris_ L.), and farmer's friend inflorescence (_Bidens pilosa_ L.). The daily and total numbers of eggs/female were higher in farmer's friend inflorescence (5.7 and 163.3 eggs, respectively) than another substrates evaluated. The results indicated the suitability of farmer's friend inflorescence as an ovipositional substrate for _O. insidiosus_ female, being the preferred one in no-choice and choice tests. Also the females showed better reproductive performance having farmer's friend inflorescence as an ovipositional substrate.

Insect damage in young eucalypt plantations in New South Wales, Australia: 1996-2003.

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State Forests of NSW manages over 27, 000 ha of young eucalypt plantations, planted since 1994. The Forest Health Surveillance program was established in late 1995 to help protect this valuable and expanding resource. Surveys are conducted annually in the majority of these plantations. Over 75 species of insect pests were observed damaging these plantations. Herbivorous insects were by far the main damaging insects, both in terms of numbers observed and damage sustained by plantations. Christmas beetles (*Anoplognathus* spp.) and Chrysomelid leaf beetles (*Chrysophtharta* spp. & *Paropsis* spp.) were the most common and damaging insects throughout the whole period of surveillance. *Eucalyptus dunnii* and *E. grandis* sustained the most damage from these insects. *Creiis lituratus* psyllids became a major pest of *E. dunnii* from 1998 onwards, but were not reported as a pest of eucalypts prior to this. Stem borers (longicorn beetles and giant wood moths) were a major problem in *E. grandis* greater than 3-years-old. Several new species of insects were identified during these surveys. The Forest Health Survey Unit provides expertise and training in all aspects of forest health to State Forests' operational divisions, and is an integral component of State Forests' risk management strategy.

Pest status of *Cosmopolites sordidus* (Germar) (Coleoptera; Curculionidae) in the Canary Islands

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The banana weevil *Cosmopolites sordidus* currently affects all the islands of the Canary Islands archipelago. Chemical control is proving very difficult, given the tunneling habit of the weevil. Alternative control methods, such as protease inhibitors and biological control using entomopathogenic nematodes and fungi, have shown to be very promising. Studies to determine geographical origin have to date revealed several different ecotypes. Banana cultivar susceptibility is under study and a new method to evaluate infestation is being developed.

Acaricidal and insecticidal properties of a Chenopodium-based botanical

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The emulsifiable concentrate UDA-245 (25%EC, v/v), based on an essential oil extract from *Chenopodium ambrosioides* var. *ambrosioides,* a North American herbaceous plant, was compared to commercially available pesticides tested for their acaricidal and insecticidal effects on common pests of ornamental greenhouses. The products were tested with the adult stages and on egg hatch of the twospotted spider mite (*Tetranychus urticae* Koch) (Acari: Tetranychidae) and of the European red mite (*Panonychus ulmi* (Koch)) (Acari: Tetranychidae), with the adult and nymphal stages of the green peach aphid (*Myzus persicae* (Sulzer), Homoptera: Aphididae), and with the adult stages of the western flower thrips (*Frankliniella occidentalis* (Pergande) Thysanoptera: Thripidae) and of the greenhouse whitefly (*Trialeurodes vaporariorium* (Westwood) Homoptera: Aleyrodidae*).* Side effects on the adult stage of the whitefly parasitoid, *Encarsia formosa* Gahan (Hymenoptera: Aphelinidae) were also determined for the potential use of the product in integrated pest management programs using biological control agents. Results suggest that a greenhouse IPM program using UDA-245 could effectively and selectively control mite infestations by treating 'hot-spots' with negligible effect on biological control agents when treating before introduction or when natural enemies are absent.

Tart cherry IPM in the United States: on-farm trials of reduced-risk insecticides and surveys of IPM adoption

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The United States is the world's largest producer of tart cherries with approximately 75% grown in Michigan. Entomologists, plant pathologists, horticulturists, and soil scientists from Michigan State University, University of Wisconsin-Madison, and Utah State University are collaborating on a project to implement reduced-risk orchard management strategies across the US tart cherry industry. Although the Tart Cherry Orchard Management Program is wide in scope, this poster covers two major aspects of this program. The first aspect is the evaluation of reduced-risk insect management in on-farm trials in Michigan. These trials were conducted in Michigan's three major growing regions in blocks ranging from 3 to 5 hectares. Pest presence, damage, and non-target effects in reduced-risk blocks were compared to conventionally managed blocks. The second aspect of the Tart Cherry Orchard Management Program covered in this poster is results of an industry-wide grower survey. Self-reported use of IPM strategies and grower perceptions of IPM adoption challenges are presented.

Pheromones of cocoa mirids

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Several genera and species of mirids (Heteroptera: Miridae) are pests of cocoa in Africa and Asia. In West Africa, the two main species are *Distantiella theobroma* and *Sahlbergella singularis*. Previous work showed attraction of male *D. theobroma* by conspecific females during late afternoon, and we have shown attraction of male *S. singularis* by females during the night. Analysis of volatiles collected from virgin female *D. theobroma* or *S. singularis* by gas chromatography (GC) linked to electroantennography (EAG) showed two EAG-active components in very small amounts. These were identified as the diester hexyl (*R*)-3-((*E*)-2-butenoyl)-butyrate and the corresponding monoester, hexyl (*R*)-3-hydroxybutyrate. Blends of these components attracted male *S. singularis* to traps and use of the traps for monitoring and control is being investigated in Ghana. These studies are already producing new insights into distribution and seasonality of the pests, but catches of mirids are extremely variable and we are investigating whether this is due to genuine aggregation of populations or to other factors.

Attract-and-kill for *Helicoverpa*: non-target effects

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We have developed plant-based attractants for *Helicoverpa* moths that can be used as attract-and-kill in cotton and other crops like corn and legumes. We monitored the effects of these attractants on pest and beneficial species other than *Helicoverpa*. In large-scale field trials in which about 1.5% of the area was treated, we collected dead insects on rows adjacent to the treated ones, as well as on other rows up to 35m away from treated rows. Although a large proportion of dead insects were *Helicoverpa*, substantial numbers of other noctuid pest species such as loopers, armyworms, cutworms and small numbers of sphingids and pyralids were also killed. Small-scale field trials on faba beans compared the numbers and types of insects collected on rows treated with the plant volatiles alone (without the toxicant) with those on untreated rows at various distances from the treated ones, and at various time intervals after spraying. Large numbers of hemipteran pest species were collected in the treated rows, suggesting that our attractants might also work for sucking pests. In general, attractants were not significantly attractive to beneficial groups like predatory beetles and bugs, wasps, lacewings and spiders, though there was some attraction to a few species.

Use of native Michigan plants to provide resources to natural enemies

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The use of plants to provide nectar and pollen resources to natural enemies via habitat management is a growing focus of conservation biological control via habitat management. The current focus, however, is on plants not native to the area of study. There is no reason to suspect that plants native to an area cannot perform as well as non-natives that are currently recommended. Use of native plants for habitat management has several benefits. Most of the natural enemy species affected by conservation biological control are native insect species, which may be better adapted to feed on native plants. In addition, native plants are pre-adapted to the environment and its natural enemies. Use of them adds to native biodiversity and restores native communities. We compared of a set of fifty native Michigan, USA and five non-native plants for their effectiveness as resource plants for natural enemies. Insect abundance and diversity were measured at flowering plants during the 2004 growing season. Plant and flower characteristics were measured to determine which, if any, were best correlated with natural enemy visitation. This procedure allows rapid plant screening to maximize resource plant effectiveness before performing in-field trials.

Economic thresholds for sorghum midge and corn earworm on midge-resistant sorghum in Australia

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In a series of field trials the relationship between numbers of attacking insects and resultant yield loss was determined for the two major pests of sorghum in Australia, the sorghum midge *Stenodiplosis sorghicola,* and the corn earworm, *Helicoverpa armigera.* One ovipositing female sorghum midge/panicle/day reduced yield by 1.4 g, and one corn earworm larva reduced yield by 2.4 g. The relationship was used to calculate economic thresholds for each pest, based on numbers of ovipositing females, for sorghum midge, and numbers of small-medium sized larvae for corn earworm. Midge-resistant sorghums have been developed over the past decade and are now extensively grown in Australia. The economic threshold for midge on resistant sorghum is increased by a factor equal to the level of resistance of the variety. For all new varieties coming on to the market the level of resistance is determined in a cooperative scheme between private seed companies and the Department of Primary Industries & Fisheries. The relationship between corn earworm larval numbers and yield loss is the same for midge-resistant and midge-susceptible sorghum. Successful IPM of the two major pests of sorghum in Australia is grounded on quantified economic thresholds.

Farmers participatory integrated pest management in scented rice

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Scented rice, popularly known as 'Basmati' in India, is mostly grown in the north Indian states. It hosts many pests, which often cause substantial yield losses. The excessive use of pesticides in some scented rice growing areas has resulted in reduction of biodiversity of natural enemies, outbreak of secondary pests, development of pests resistant to pesticides, pesticides induced resurgence and residue problem in grains. In this context, Integrated Pest Management (IPM) is highly relevant. IPM validation has been carried out during *Kharif* (May-October) of 1999 to 2003 in a village Shikohpur in Uttar Pradesh, where farmers solely depended on the use of intensive pesticidal applications, over an area of 40, 120 and 160 ha during 1999, 2000 and 2001 respectively. Adoption of IPM approach (seed treatment, regular pest surveillance and monitoring, conservation and augmentation of natural enemies, need based application of pesticides and some improved crop management practices) led to consistent higher yield levels of 5.69 (1999), 5.80 (2000) and 5.74 t/ha (2001) in IPM, compared to 5.03, 4.82 and 4.56 t/ha for the same years in the farmers practices (non-IPM) with reduced input cost and high cost-benefit ratios. During the subsequent years (*Kharif* 2002 and 2003), majority of the farmers continued to follow IPM tenets and got enhanced yield and economic returns. This outcome emphasizes the need to further spread the message of IPM in scented rice growers.

Reducing pesticide inputs in nurseries through the use of a portable hot water re-circulation immersion system

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As part of our Integrated Pest Management efforts to provide an improved method for nursery managers to control pests on plant cuttings, we have devised a non-chemical, hot water re-circulation immersion system for killing insect pests such as spider mites, broad mites, scale insects, mealybugs, lace bugs, and aphids on woody and herbaceous propagation material. Research has been conducted to determine the optimum temperature and duration of time for several plant species to prevent phytotoxicity and effectively kill insects. Four Maryland, USA nurseries cooperated with this project. Success of this project was determined by the efficacy of the hot water immersion method at suppressing pest populations and by monitoring the adoption of this alternative insect pest treatment system by commercial plant propagators.

The pest manangment of screw bean seed pest *Caryedon serratus* (Col.:Bruchidae) on base biological control in Bushehr province.

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Prosopis cineraria (L.) Druce is a beautiful native plant that has an important role in the life and ecological conditions in Boushehr province. *Caryedon serratus* is the most important pest on *Prosopis cineraria* because it causes the most damage in coastal areas from Kangan to Bandar-abbas. I observed oviposition to occur with mostly solitary eggs being laid and less frequently in a group (3-5 egg) on seeds and pod surfaces. After hatching, larva penetrated the seeds and destroyed them. Then, became pupa in the seeds and adults made exit holes on surface fruits. The pupal parasitoieds of Encyrtid family isolated from this pest. In 1991, selected one hectare (10000m^1) of *Prosopis cineraria* forests and determined the best method for pest control. to pay attention to wide dispersal of forests , support and survival of environment and no-use or low-use of pesticides and emphasis on biological control by support of parasitoids from Encyrtid family , integrate pest manangment (IPM) performed with following methods and the pest *C. serratus* was controlled successfully.1- the collection of seeds and pods in june and November ,isolation of seeds and disinfection by Diazinon %60 or Sevin. 2- grazing to destroy infected seeds and pods. 3- the cutting of canopy around the *Prosopis cineraria* to destroy overwintering and summering of pest. 4-only two times spray with Malathion %75 or Diazinon %60 after falling flowers and beginning of pods for support of parasitoids.

Efficacy and ecological response to pseudostem and pheromone trapping of *Cosmopolites sordidus* (Germar) (Coleoptera: Curculionidae) in South Africa

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The dominant insect pest of bananas in South Africa is *Cosmopolites sordidus*. In developing an integrated pest management strategy for *C. sordidus*, different trapping methods were compared in banana plantation field trials during autumn, spring and summer along the south coast of KwaZulu-Natal. Pseudostem traps, two different pheromone lures and control traps (pitfall traps without pheromone lure) were evaluated for their efficacy in collecting adults over five weeks. A randomised block design was used for all trials. Pitfall traps contained diluted ethylene glycol, where attracted beetles were drowned. Pseudostem traps were covered with mulch to delay desiccation and replaced once a week during surveys. Dissected adults were sexed and the number of eggs recorded. Pheromone traps collected significantly more beetles than the pseudostem traps, which were significantly more effective than the control. Significant differences in sex ratio were observed, where pseudostem traps attracted about equal numbers of males and females while pheromone traps showed a female bias. Females attracted to pseudostem traps were usually more fecund than females attracted to pheromone traps. The efficacy of pheromone traps and their low labour requirements shows promise as a monitoring tool and possible control strategy against the banana weevil in South Africa.

Entomological diversity in carrot fields in Belgium and impact of close habitat

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This work aims to characterise the entomological diversity in carrot crop (*Daucus carota* L.) in South Belgium. During years 2000 and 2001, 271.083 flying insects were caught in 24 fields using yellow water traps. 133 insect families were identified. Results show that insect population diversity and stability increase with crop development. Aphid problems occur only at the beginning of the season, when young carrots are especially susceptible. When crop reaches maturity, beneficials control prevents any aphid infestation. We also asses the impact of vegetation close to fields: other crops, herbaceous strips hedges and woody borders. Results show also that agro-environmental measures enhance insect density and diversity, mainly for beneficial guilds. Results are discussed in relation to potential effects on aphid infestations.

Peelminer growth and development in California Citrus

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Peelminer, *Marmara gulosa* Guillen and Davis (Lepidoptera: Gracillariidae), has been a persistent pest of citrus fruit in the San Joaquin Valley in central California since 1999. Peelminer caused widespread and extensive damage in a short period of time and was of major concern to growers being only one of four species to directly attack the fruit. Studies were initiated on three different varieties (grapefruits and navels) to determine growth and development, distribution, abundance, and feeding habits of peelminer in commercial settings. Larvae extensively mine fruit epidermis creating serpentine mines of up to 50 cm over the course of their development. Peelminer populations are multivoltine in the San Joaquin Valley and they overwinter as larvae on citrus fruit and stems as well as stems of several other host plant species. Peelminer populations expanded through the valley rapidly; within commercial citrus plantings, populations were typically localized upon initial discovery and spread throughout the grove within a single season. Peelminer populations were extremely high on fruit subsequent to its discovery and remained high with a range of 0.17 to 0.43 mines per fruit in grapefruit and 0.07 to 1.17 mines per fruit in navels in the 2003 season.

Triphelio - An international research project for sustainable control of lepidopterous pests in olive groves.

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The olive moth as well as the jasmine moth are serious pests on olive. The EU-funded project TRIPHELIO (Contract ICA4-CT-2001-10004) explores biological and biotechnical methods for control of these pests by:

Evaluating pheromone-based mating disruption in different regions and commodities of olive cultivation.
Developing strategies for enhancement of natural enemies in the olive ecosystem.
Exploring inundative releases of *Trichogramma*-wasps as control method.

Promising effects were found in current field trials of mating disruption as well as of enhancing vegetational diversity in the olive grove ecosystem. Concerning *Trichogramma*-releases, further improvement of their efficacy is needed, for instance by the use of local, environmentally well adapted *Trichogramma*-species.

Monitoring the rice leaf bug, *Trigonotylus caelestialium*, with synthetic sex pheromone

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The rice leaf bug, *Trigonotylus caelestialium* (Heteroprea; Miridae), is widely distributed across Japan. The adults and nymphs of this stink bug attack the heads of rice plants and cause pecky rice. In 1999, the stink bugs had inflicted serious damage on rice plants in the northern part of Japan. It is well known that the females secrete sex pheromone to attract the males. The sex pheromone components of this stink bug have been reported. We made an attempt to employ the synthetic sex pheromone for population monitoring. Water pan trap baited with a rubber cap containing synthetic sex pheromone was used for monitoring the seasonal prevalence of the occurrence of adult stink bugs. The trap baited with 0.01mg synthetic sex pheromone attracted many males and was nearly equivalent to a trap baited with 10 virgin females. The attraction period of the lure was more than one month. These results suggested that the pheromone trap can be used more than one month as a tool for monitoring the population of stink bugs in the fields.

Effect of suppressing density for green rice leafhopper, *Nephotettix cincticeps*, on resistant near-isogenic lines and response of local populations to resistant varieties

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Use of crop resistance has the advantages of environmental load reduction, reduced labor and lower costs and is an effective method for integrated pest control. We developed four types of near-isogenic lines (NILs) of the Kinuhikari variety of rice with different resistance genes for green rice leafhopper (GRH). The fact that the response to resistant varieties differs for local populations of GRH is known. Thus, GRH populations were collected from four locations in Joetsu, Mito, Chikugo, and Kagoshima and virulence to each NILs was investigated. The result was that it was lower in the Joetsu and Mito populations to lines having resistance genes Grh 1 and Grh 2. In contrast, virulent individuals were present for the Chikugo and Kagoshima populations on the order of 30-40 percent. With line having a combination of Grh 2 and Grh 4, the proportion of virulent individuals was lower for all populations. In addition, resistant NILs were cultivated in paddy fields in Joetsu and Chikugo and the effect of suppressing GRH density was examined. The result was that the density of GRH on each NILs was low in both Joetsu and Chikugo and a strong effect on suppressing density was noted.

Leafminers, *Liriomyza* spp. (Diptera: Agromyzidae), in Vietnam: distribution, biology and insecticide resistance

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Liriomyza sativae is the main polyphagous *Liriomyza* species appearing as a pest in vegetables in most of Vietnam. *Liriomyza huidobrensis* is a major problem in the highland area around Dat Lat in southern Vietnam. In addition, *Chromatomyia horticola*, *L. bryoniae* and *L. trifolii* are present, but their pests status on vegetable are not clear. Parts of the biology of *L. sativae* have been studied, and the native parasitoid complex seems to be important. *L. sativae* was collected from four locations in the Hanoi area and tested against five insecticides commonly used against leafminers in northern Vietnam. Resistance towards cypermethin was found in the three populations from locations with intensive insecticide use.

Effect of *n*C24 agricultural mineral oil on two-spotted mite (*Tetranychus urticae* Koch) and cotton aphid (*Aphis gossypii* Glover)

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An *n*C24 agricultural mineral oil was evaluated under laboratory conditions for its bioefficacy against adult female two-spotted mite (Acari: Tetranychidae) and cotton aphid (Hemiptera: Aphididae) on cotton leaf discs. Sprays of water only, and aqueous emulsions that gave 20, 40, 60, 80 and 100 mg oil cm^{-2} deposits, were compared. All sprays were applied with a Potter spray tower. The impacts of sprays on arthropod mortality were assessed 24 h after sprays were applied to infested leaf discs. The impacts of spray deposits on oviposition by *T. urticae* females and nymphs produced by *A. gossypii* females were determined 24 h after release of the adult female arthropods onto dry spray deposits. All emulsions applied to adult mites and aphids led to significant mortality and mortality increased with dose. All emulsions significantly reduced oviposition by mite females but deposits < 60 mg cm^{-2} had no impact on numbers of aphid nymphs recorded on the leaf discs. The results are discussed in relation to recent field studies on cotton and other crops.

The 'tool kit' for extending IPM information for diamondback moth control to the Brassica industry of Australia

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A range of tools has been selected to provide what is commonly called 'technology transfer' or 'extension' to Brassica growers and their advisors in Australia. The range comprises methods that are face-to-face, electronic, direct-mailed, and voluntarily collected. The subjects covered via these methods are tactics of IPM including: Biological - such as identification of natural enemies and pests, Managerial - such as monitoring and buying healthy transplants, Chemical - advocating the use of insecticides as a last resort and 'soft' option insecticides such as Bt's, as well as highlighting the use of the insecticide resistance management strategy (IRM) developed for Diamondback moth (DBM) specifically, and Genetic - cultivar selection based on research findings. Some examples of the tools include: Workshops and field education that includes selected local and overseas invited speakers. A national Brassica IPM newsletter informs about the latest in pest and disease management. A handbook of DBM management contains updated modules about the pest and the various IPM tactics recommended. Direct education about IRM to staff at chemical stores ensures the same level of knowledge across industry. Website information and downloadable crop sampling plans are yet another tool that is continuing to move the industry towards IPM practices.

Effect of soil fertility amendments on crop character and infestation by cabbage pests

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Field studies were conducted to assess the effect of market crop wastes (MCW) derived soil fertility amendments vs. a conventional fertilizer on crop character and population dynamics of aphids (*Brevicoryne brassicae* and *Myzus persicae*) and the diamondback moth on cabbage, *Brassica oleracea* var. *capitata*. Treatments were 1) MCW compost incorporated in the soil, 2) un-composted MCW incorporated in the soil, 3) Un-composted MCW applied as a surface mulch, 4) a conventional chemical fertilizer (NPK) incorporated in the soil, and 5) the untreated control. Response variables recorded included shoot length/width (cm), leaf area, leaf toughness, %tissue nitrogen, aphid infestation ratings, and diamondback moth counts. Results indicated that for growth parameters, plots receiving amendments did slightly better than the untreated ones, also, untreated plots had the lowest infestation of the diamondback moth.

Effect of *Eucalyptus* and *Tylophora* extracts on feeding of *Helicoverpa armigera* (Hübner) larvae.

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Helicoverpa armigera is an important pest of many crop species. Environmentally benign strategies are required to manage this pest. Hexane and ethanol extracts of *Eucalyptus camaldulensis* and *Tylophora indica* leaves were evaluated for their antifeedancy against *H. armigera* larvae. Cabbage leaf discs (12.5 sq cm) dipped in the test solution for five seconds were used for feeding the larvae. Four fifth instars, starved for 4h, were fed on the treated cabbage leaf discs in eight replicates for 24h. Feeding values were calculated in comparison to solvent treated control. The calculated percent feeding inhibition values were analyzed by Probit analysis to calculate the EC_{50} values. Ethanol extracts of both the plant species exhibited high feeding inhibition activity having EC_{50} values of 6.92 and 2.81% for *Eucalyptus* and *Tylophora*, respectively as compared to hexane extracts which recorded the EC_{50} values of 9.28 and 5.21%. Feeding inhibition when computed against different concentrations showed a high exponential growth (R-sq change) indicating the linearity of the equation and sensitivity of the insect to different concentrations.

Predicting spring flights of *Etiella behrii* (Lepidoptera: Pyralidae): a pest of lentils in southern Australia.

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The Lucerne seed web moth, *Etiella behrii* (Zeller) (Lepidoptera: Pyralidae), is a serious pest of lentils in southern Australia. The larval stage of *E. behrii* bores into immature pods to feed on developing grain, resulting in yield losses and a reduction in grain quality. Current Australian receival standards for lentils allow no more than 1% damage by insects. Producers' ability to supply grain of this quality relies on the effective management of *E. behrii.* Insecticide treatments often fail to provide adequate control of *E. behrii* in lentil crops. We suspect a primary cause is the inappropriate timing of many of these treatments. Monitoring the activity of moths and timing spray applications to target peak adult activity prior to egg laying may prove to be an effective management strategy. To identify the seasonal occurrence of *E. behrii* activity we have analysed the relationship between the timing of spring flights in historical light-trap records and temperature. Degree-days have been used to predict the phenology of *E. behrii* and assist in targeting pesticide applications, thus furthering the development of integrated pest management procedures for this pest.

Efficacity of pheromone rates on male captures of potato tuber moth *Phthorimaea operculella* (Zeller)(Lepidoptera: Gelechiidae).

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Potato tuber moth (PTM), *Phthorimaea operculella* (Zeller) is one of the most serious insect pests of potatoes both in the field and in storage in regions with hot, dry summers throughout the World. Larvae develop on potato foliage and in tubers causing direct losses of edible product. Chemical treatments to control this pest have shown limited efficacy, especially when larvae are in the soil. The use of synthetic pheromones as a tool to interfere with mating has been widely demonstrated in numerous moths and in other insect species. During the summer season 2003-2004, a study was carried out at Valle del Elqui, IV Región, Chile, to evaluate the efficacy of different rates (0.2; 0.5; 1; 2 and 5 mg) per trap of PTM sex pheromone mixture (Trans-4, cis7-tridecadieno-1-ol.acetato and Trans-4,cis7,cis10 tridecatrieno.1.ol-acetato, at rate 1:1,5) on PTM male captures. The results showed that the pheromone capsules loaded with lower rates (0.2 and 0.5 mg.), captured higher numbers of male moths per trap. Mass trapping technique studies will be continued to optimize the rate of pheromone per trap in order to offer a non-chemical and environmentally safe method of control of PTM.

The efficacy of coloured pan traps in sampling local and distant hover-fly (Diptera: Syrphidae) populations.

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Coloured traps are frequently used to attract and catch insects, such as in the monitoring of populations of beneficial insects in 'classical' or 'conservation' biological control. They are also used in the evaluation of the recovery of insect populations after disturbance. However the fact that traps may be visible to the insects over a considerable distance can influence the interpretation of catch data. In this work, traps consisted of two-litre plastic containers which were filled with water, detergent and a preservative. They were placed in a randomised block design in a 30m × 50m area of bare soil. The treatments were: inner and outer containers yellow; inner container yellow and outer green; both containers green. The hypothesis tested was that completely yellow traps would attract flies from a distance, while yellow/green traps would catch flies from above. Green traps were controls. *Melanostoma fasciatum* was the commonest species in each treatment, with twice as many captured than *Melangyrna novaezelandiae*. For both species, the largest number of hover flies was trapped in completely yellow traps, an intermediate number in green and yellow traps and the lowest number in green traps. It is suggested that if hoverfly numbers relating to a particular distance along a transect are required, consideration should be given to the ability of hoverflies to see yellow traps from a distance. Traps, which are green outside, would be more appropriate to reflect the local abundance of hover flies.

Managing soil insect pests of potatoes in Australia

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Soil insect pests consisting of a range of species of beetles and one species of aphid, the rice root aphid (*Rhopalosiphum rufiabdominalis*), have the potential to reduce yield and quality of Australian potatoes. A national collaborative project in the major potato growing regions is studying methods of monitoring these insects so farmers can introduce pest reduction practices on a needs basis. Some of the soil pests have not been identified, so a second objective of the project is to expand the known list of soil insect species that can affect potato crops. To date, monitoring methods involving pre-crop soil sampling for whitefringed weevil (*Naupactus leucoloma*) and African black beetle (*Heteronychus arator*) have proven to be of high order reliability in providing advice to farmers whether treatment is required to protect crops. For African black beetle, light traps are a useful warning tool for pest invasions of growing crops. For potato wireworm (*Haptesus hirtus*), baiting with potato tuber pieces has been shown to be an effective method of assessing insect distribution and abundance. The main insects whose identification is yet to be clarified include cockchafers and wireworms. We hope to reduce the pest status of soil insects in Australia's potato crops by producing a guide for farmers and crop scouts on pest identification and monitoring methods.

Occurrence, infection and research advancement of the vegetable leafminers in China

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Liriomyza sativae and *L. huidobrensis* are two serious pests among four species of leafminers infested vegetables in China. Our investigation showed that their host plants infer to more than 20 families, 100 species, even including barley and wheat. The biology, such as adult eclosion, emergence from soil, feeding and oviposition, migration and dispersal, larval feeding and so on, of both *L. sativae* and *L. huidobrensis* was investigated systematically. Effects of temperature on development, mortality, fecundity and longevity of the two leafminers were examined in the laboratory. Overwintering and cold-hardiness of the pests, which provided basis for evaluating northern limit of overwintering, were also studied. The results showed that both *L. sativae* and *L. huidobrensis* had high productivity and strong adaptability to the environment. A preliminary study on diversity of feeding habits of *L. huidobrensis* revealed that the activities of carboxylesterase and acethylcholines increased with the adaptation of *L. huidobrensis* to wheat. We also investigated the parasites of the vegetable leafminers in China and found more than 30 species of parasites, of which the eulophid and braconid wasps are the main species. According to our study results we designed an integrated control protocol for the vegetable leafminers in China.

Influence of vineyard floor vegetation on the abundance of winged natural enemies

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According to Conservation Biological Control, vineyards with vegetated floors should have greater numbers and performance of natural enemies than those with cleared floors because more resources like food and shelter are available. The influence of vegetated and cleared vineyard floors on winged natural enemies is being studied by evaluating sticky trap catches of insects at floor and canopy levels. Initial results show that both numbers of species and total numbers of potential natural enemies of grapevine pests were significantly higher in traps on a vegetated floor than a cleared floor. Canopy catches were significantly lower than floor catches and unaffected by floor treatment. Trap catches of most wasp species and the ladybirds *Diomus notescens* and *Hippodamia variegata* were significantly higher from the floor than in the canopy. Catches of the ladybirds *Harmonia conformis* and *Coccinella transversalis*, and several wasp species were significantly higher in canopy traps than floor traps. Inter-row vegetation increased numbers of natural enemies in vineyards but natural enemy activity was concentrated on the floor and not in the canopy indicating that commensurate increases in grapevine pest control are unlikely. Differences in species composition between canopy and floor traps highlight different patterns of flight and resource use by natural enemies.

Pheromones of *Crocidosema plebejana* (Lepidoptera: Tortricidae), *Creondiates dilutus* (Hemiptera: Miridae) and *Earias huegeliana* (Lepidoptera: Noctuidae),

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Extensive cotton growing areas in Australia are occasionally affected by pests like *Crocidosema plebejana*, *Earias huegeliana* and *Creontiades dilutus*. We have identified the major sex attractant pheromone components for these species from effluent air surrounding calling females, and pheromone gland contents, by GC-MS. We conducted studies using Delta traps arranged in Latin Square designs to determine optimum attractive blends for all three species. This poster will compare the results with closely related species from other countries and discuss the prospects of using the optimised blends to monitor and forecast outbreaks of these pests

Technology development for fruit fly detection and exclusion programs

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Fruit flies in the family Tephritidae are among the most destructive insect pests in the world. In countries where pest fruit flies are present, the negative economic impact results not only from the direct costs of crop damage and control activities, but also from limited access to market agricultural commodities in non-infested countries. Fruit fly detection and exclusion programs in the United States are supported by the United States Department of Agriculture, Animal and Plant Health Inspection Service, Center for Plant Health Science and Technology (CPHST). An overview of the technology development activities of CPHST laboratories in Florida, Guatemala, Hawaii, and Texas will be presented. These activities include: 1) technology for Mediterranean and Mexican fruit fly suppression programs that improve mass production and enhance field performance of males in the sterile insect releases; 2) integration of biological control and sterile insect releases for suppression of the Mediterranean fruit fly; 3) validation of new trapping and bait station technologies for APHIS, state and international fruit fly detection programs; and 4) molecular diagnostics for determining the population origin of exotic fruit flies detected in the United States.

Behavioural control of the boll weevil through BWACT in cotton farms in the northeast of Brazil

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The boll weevil, *Anthonomus grandis*, is the most important pest of cotton in the Brazilian Northeast and could cause significant losses in the production. The Boll Weevil Attract and Control Tube (BWACT) have been used by the producers as tactic of behavioural control. Because of their located and specific control, this method is considered an appropriate tool to the concepts of Integrated Pest Management (IPM), working as alternative to the chemical control. This study sought to evaluate the use of such devices in the control of the boll weevil in areas of cotton farms in the Brazilian Northeast. In comparative experiments among conventional management (where the use of the chemical control prevails), IPM, and their interactions with the use of BWACT, smaller number of applications of insecticides was verified for the control of the insect in areas in that it was allied MIP plus BWACT. The device was efficient on the control of the boll weevil when installed at the planting time. The BWACT reduced remaining populations of the insect after the stalk destruction. The adoption of IPM plus BWACT resulted in larger liquid incomes than the non adoption of IPM and the non use of the device..

Recording the occurrence of the bean bug by an automatically counting trap, "Altered Mushidas", baited with synthetic attractants of the bugs

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The bean bug, *Riptortus clavatus* (Thunberg) (Heteroptera: Coreidae), is one of the major soybean pests in Japan. Since the adults invaded into the soybean field, monitoring of their movement is essential to control them. A *Mushidas* is an automatically counting trap equipped with wireless communication tools, originally developed for monitoring the occurrence of the lepidopreran pests baited with their synthetic sex pheromone. We improved the *Mushidas* attaching the collision board in the upper part of its body so as to catch the bean bug effectively by using synthetic attractants of the bug. The whole body was painted in green because a preliminary experiment showed that numbers of other insects caught were significantly fewer in the green trap than those in the white and yellow ones. Three sets of the trap were installed in the soybean field, and they were proved to continuously work for about 5 months with the records at an interval of 15 min. It was found that the most of the bugs were caught in the daytime, having no clear peak. Those traps might be able to monitor the occurrence of the bugs earlier than the conventional water pan traps.

The biology and management of the Madeira mealybug, *Phenacoccus madeirensis*

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The Madeira mealybug has become a major problem on greenhouse grown ornamentals with control failure in many greenhouse operations. Experiments were conducted to determine the developmental biology of Madeira mealybug and efficacy of most available insecticides against this pest. Using chrysanthemum as the host plant, the total duration of development of Madeira mealybug was 66 days at 15°C, 46 days at 20°C, and 30 days at 25°C. More than 75% of eggs completed development to adult. The proportion of females in a given population was about 0.5. The insecticides acephate and bifenthrin and the growth regulators kinoprene and buprofezin where the most efficacious. The natural enemies, *Anagyrus loecki* and *Diomus austrinus*, were explored as potential biological control agents. The biggest management problem was associated with good spray coverage and the behavior of the insect to hide in difficult to reach places on the plant and to move to the pots and benches to hide.

Pest management in Honduran subsistence maize production: contribution of training, agro-landscape composition and region-specific pest pressure

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A major pest in Honduran maize production is the fall armyworm (FAW), Spodoptera frugiperda. To manage pest outbreaks, small-scale farmers tend to rely on unsustainable practices, including improper use of pesticides. In the past, various institutions have conducted IPM extension programs to alter farmers' pest management. Despite those efforts, a formal impact assessment yet needs to be conducted and obstacles for IPM adoption await identification. IPM packages for subsistence agriculture consist partly of manipulating the pest's natural enemies. Since most natural control agents have ecological requirements beyond the field edge, success of IPM largely depends on the agricultural landscape. We assessed IPM adoption through farmer surveys. Surveys were conducted in communities with differing IPM training history and pest pressure. We evaluated farmers' agro-ecological understanding, knowledge and its influence on pest management behavior. Secondly, we quantified opportunities for efficient FAW management existing within the agro-landscape. Pest pressure and natural control were linked to the composition of the agricultural landscape and back-crossed to farmers' pest management. Farmers base their pest management on an overall appreciation of agro-ecology and FAW pest-pressure. Farmers' agro-ecological understanding is influenced by perceptive experience and traditional knowledge. Agro-landscape composition strongly affects the assemblage of generalist predators and parasitoids responsible for natural FAW control.

Integrated pest management to control California red scale *Aonidiella aurantii* (Maskell) in the Mediterranean area (Sardinia, Italy)

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In Sardinian citrus groves, the use of *Aphytis melinus* DeB. and *Chilocorus bipustulatus* (L.) alone has not allowed to keep infestations of California red scale *Aonidiella aurantii* (Maskell) under the economic threshold. The use of a prediction phenological model, based on growing degree-day accumulation between the peaks of males flight and the highest density period of crawlers (100 and 250 °D accumulate in temperatures between 11.7 °C and 37.8 °C), allowed us to determine the optimal time for effective applications, to reduce chemical treatments and to apply low environmental-impact insecticides. In addition, specific surveys on within-tree spatial distribution of California red scale on orange trees were conducted, to determine the most infested parts of the canopy from which the insect spreads throughout the tree. Pruning the heavily infested twigs allowed to reduce the centres of infestation of *A. aurantii* population, to create unfavourable microclimatic conditions for population development and to reduce population density using low environmental-impact insecticides. These integrated control strategies were effective for both California red scale infestation control and protection of its natural antagonists *A. melinus* and *C. bipustulatus*. Research conducted in the MiPAF plan "Search and experimentation in the Italian citrus cultivation" publication

Sirex noctilio in Chile operational program for detection and control in the Arauco companies

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Sirex noctilio was detected close to commercial plantations of *Pinus radiata* in Chile by 1999. This regulated pest is being monitored by the Plant Health Agency (SAG) over affected areas in the south of Chile. Radiata pine supports the forest industry of Chile, with US$ 2.218 million of total export in 2003. The government and private companies have now implemented a program for the early detection of *Sirex* over previously unaffected areas. Arauco is the largest forestry company in Chile, with 500.000 ha of Radiata pine plantations. About 370,000 ha are considered to be at risk to *Sirex* attack. Stands of Radiata pine are being classified in different levels of risk to *Sirex* establishment by means of a GIS system. The criteria to define stand susceptibility to *Sirex* were: silvicultural management, site index, age, average diameter, density, and distance of declared focus. The information generated by the GIS analysis was used to assist trap tree plots placement in Arauco plantations. A total of 340 trap tree plots were established over the main risk stands thus far. These plots included trials with different herbicides (Dicamba, Tryclopyr and Gliphosate), with aims to optimize the trap tree system in Chile.

Effects of regulatory pressures on pesticide use in New Jersey peach production

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New Jersey peach growers regularly participate in a cooperative extension fruit integrated pest management program. Grower pesticide use records are annually collected from program participants. In 1997 an Access database was created as a repository for all IPM data. Pesticide use was analyzed for 1998 through 2003. All participating farms are profiled within the database, permitting accurate records of acreage, cultivar, and location. A 'back end' data file was created listing all pesticides used by material (trade) name, active ingredient (ai), % ai, chemical class, function type, and a standard retail price. Queries were designed to generate individual grower reports and analyze area and statewide pesticide use. Queries analyze by material name or ai, and track total use per acre, rate per acre, number of applications used, and the % of total crop acreage the pesticide was used on. In 1998 organophosphates (OP's) esp. azinphos-methyl (AZM) were the most commonly used materials, while synthetic pyrethroids (SP) showed only minor use. By 2003 the number of AZM applications decreased by 30%, while SP applications more than doubled. Changing trends were also seen with other materials, especially oil, neonecotines, and other OP's.

IPM of *Tanymecus dilaticollis* on maise in Romania

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Maize, cultivated over three million ha is one of the most important crops in Romania. The crop technology involves complex protection measures vs. pests and pathogens. The greatest damage is produced by *Tanymecus dilaticollis* attack even from the emergence. In order to avoid this damage or total crop spoiling, the adoption of a complex system based on seed treating is necessary, as follows:

- for areas where the preceding crop was a winter spike cereal, vetch-oats mixture, bean, soybean, pea or linseed, the chemical treatments are not indicated, because the density registers low values till 3 individuals/m^2;
- if the preceding crop was maize, sunflower, sugar beet or sorghum, the maize seed treating is recommended with the following ingredient: carbofuran (8,750 g.a.i/t seed), thiamethoxam (3,150 g.a.i/t), imidachloprid (4,800 g.a.i/t), bensultap (12,500 g.a.i/t).
- in the case in which, after plant emergence, there are favourable climatic conditions for pest appearance, the application of one correction treatment during vegetation is recommended on seed areas treated with: thiamethoxam (25,0 g.a.i/ha), t

A research team approach to solving strawberry pest problems in Florida

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Florida produces winter, fresh-market strawberry fruit on about 2800 ha. valued at over $170 million annually. The fruit are produced in a environment hosting numerous damaging arthropod, nematode, and weed pests and diseases including twospotted spider mite (*Tetranychus urticae* Koch), melon aphid (*Aphis gossypii* Glover), strawberry root aphid (*Aphis forbesi* Weed), fall armyworm (*Spodoptera frugiperda* J.E. Smith), southern armyworm (*Spodoptera eridania* (Stoll)), nitidulid sap beetles, *Drosophila* sp. fruit flies, *Frankliniella* spp. thrips, root knot nematode (*Meloidogyne hapla* Chitwood), sting nematode (*Belonolaimus longicaudatus* Rau), gray mold (*Botrytis cinerea* Pers.), powdery mildew (*Sphaerotherca macularis* (Wallr. Ex Fries) Jacz.), anthracnose (*Colletotrichum* spp.), angular leaf spot (*Xanthomonas fragariae* Kennedy and King) and numerous weeds. The University of Florida has assembled an interdisciplinary team of entomologists, nematologist, plant pathologists, weed scientist, and a breeder who are developing and refining integrated approaches for management of these pests. The team approach will be presented.

Weeds and native vegetation as hosts for pest and beneficial thrips (Thysanoptera) in a horticultural landscape

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Revegetation programs have not progressed in the main horticulture-producing region of South Australia because of perceptions that native plants harbour pest thrips of horticultural plants. To determine the distribution and abundance of thrips, we sampled flowers from twenty-two species of weeds and twelve species of native plants at monthly intervals for a year. *Frankliniella occidentalis* was found on all species of weeds sampled throughout the year. *Thrips tabaci* and *F. schultzei* were found on most species of weeds. At least one of these thrips was more likely than not to be found on weeds in the Brassicaceae, Solanaceae, Malvaceae or Zygophyllaceae. However, these pest thrips were only found on four species of native plants, and infrequently. *Thrips imaginis*, a native thrips, was found on all species of weeds and native plants sampled, but was more likely to be found on plants greater than 100 m from crops than on plants less than 10 m from crops. *Haplothrips victoriensis*, a predatory thrips, was most frequently found on weeds in several plant families and rarely on native plants. These results suggest that pest management can be improved by replacing weeds with native plants that do not harbour pests of horticultural crops.

Overwintering of *Heliothis virescens* (F.) and *Helicoverpa zea* (Boddie) (Lepidoptera: Noctuidae) in cotton fields: implications for pest and resistance management

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Following the 1995-2001 crop seasons, population densities of pupae of tobacco budworm (TBW), *Heliothis virescens* (F.), and corn earworm, *Helicoverpa zea* (Boddie), overwintering in cotton fields in Monroe County, Mississippi, USA, were estimated by digging a total of 43,542 m-row. The total, area-wide TBW population emerging from all overwintering sites was estimated from catches in pheromone traps whose absolute efficiency is known. Before introduction of *Bacillus thuringiensis* Berliner (*Bt*)-transgenic cotton in 1996, cotton fields are estimated to have typically accounted for less than 10% of the total, overwintered TBW population. During 1996-1999, cotton fields accounted for less than 2% of overwintered TBW. During 2000-2001, the contribution of cotton fields to overwintered TBW remained low despite a dramatic increase in use of herbicide resistant (*HR*)-transgenic cotton cultivars starting in 2001. Consequently, preemptive tillage of cotton fields to kill overwintering TBW pupae would not appreciably suppress in-season population densities of this pest. However, given that movement is short-range during the crop season and fall generations, overwintering TBW pupae resistant to insecticides or counteradapted to host plant resistance traits [e.g. (*Bt*)-transgenic cotton] should be concentrated in cotton fields. Failure to destroy these pupae by tillage would reduce the time to control failure.

Managing *Helicoverpa* spp. on cotton with semio-(signalling) chemicals

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This study is focused on the non-volatile chemical components of organ surfaces of cotton and alternative host plants that influence acceptance or rejection of such plants by adults and larvae of *Helicoverpa* spp. Solvent extracts of both leaf surfaces and whole leaf homogenates of three cotton genotypes, as well as plants used as trap and refuge crops in *Helicoverpa* spp. management, were assessed for bioactivity towards both adult and larvae of *Helicoverpa* spp. in the laboratory. Different specific bioactivity properties have been demonstrated in crude extracts of the various plants. Bioassay-directed fractionation of the extracts using solid phase extraction methodology was followed by analysis and compound isolation by HPLC, with a view to identifying particular compounds responsible for the bioactivity. A leguminous plant, herein referred to as Plant X, has shown oviposition and feeding deterrence towards *Helicoverpa* spp. Two potential deterrent chemical compounds in Plant X have been isolated and identified. Most of the active fractions in all plants contain compounds showing UV spectroscopic properties characteristic of flavonoids and/or flavonol glycosides. We conclude that natural chemical tools for the modification of insect behaviour could be an important tool in an integrated pest management program targeted at *Helicoverpa* spp. in cotton.

The tobacco budworm, *Heliothis virescens* (F.), in North Carolina tobacco: decommissioning a prominent pest?

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The tobacco budworm, *Heliothis virescens* (F.), has long been regarded as a key pest of tobacco in North Carolina and other areas in the southeastern United States. This insect causes at least two main types of damage: defoliation of expanding leaves (the far more common type of damage), and destruction of the apical meristem of the plant. Historically, the threshold for the tobacco budworm in North Carolina has been 10 % infested plants. Flue-cured tobacco crops in the eastern part of the state have averaged about 1.5 insecticide applications a year for control of this insect. A growing body of evidence assembled over the last 7 years suggests that the tobacco budworm's ability to compromise tobacco yield and quality in North Carolina is far lower than previously thought. The critical form of damage is destruction of the apical meristem, which is a relatively rare event in the field. In light of this evidence, the threshold for this insect will likely be raised substantially. This action should substantially reduce insecticide use in North Carolina tobacco.

Flight pattern of the overwintering generation of *Archips podana* (Lep.: Tortricidae) based on data from pheromone traps

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In 1993 – 1999 and 2002 – 2003 the flight activity of *Archips podana* was investigated by pheromone traps placed in four apple orchards in Central and East Bohemia. The course of the flight activity of *A. podana* was expressed in relation to calendar dates and biological time (degree-days) and the effects of pest control strategies and weather conditions on total catches were determined. The cumulative catches of *A. podana* males were plotted against time of the catch expressed in sum of degree-days (DD) above 10 °C and approximated by Richards´ function. The common parameters of Richards´ function could be found for the overwintering generation of *A. podana* from all localities. The reasonable predicting of the beginning, the peak and the end of the flight activity of the overwintering generation of *A. podana* population in Central and East Bohemia was ascertained. The flight activity of *A. podana* started at 200 DD (5% catch), median catch (50% catch) was at 320 DD and the flight ceased at 420 DD (85% catch).

Arthropod pest management in Christmas trees in the Pacific Northwest

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Several arthropod pest species attack commercial Christmas trees in the Pacific Northwest United States. We have been evaluating various control agents for several pests that Christmas trees. In particular, we have focused on root aphids, adelgids and eriophyid mites. The insecticide, Aphistar has been found to be a particularly effective control for root aphids. Aphistar is applied to foliage and when applied at a rate of 8 oz. product /acre, controls these root pests for up to 6 weeks on Noble fir. A new insecticide, Acetamidprid was evaluted for control of adelgids. Acetamidprid applied to foliage at rates of 0.05 lb ai/100 gallons water and 0.1 ai/100 gallons water provided excellent control of adelgids on Douglas fir. Thiodan and Mesa were evaluated for control of an eriophyid mite, *Nelepella longoctonema* on Noble fir. Thiodan was the most effective product, but Mesa also provided control of this species. Although we have focused on pesticides for control of Christmas tree pests so far, our goal is to develop integrated pest management for pests attacking Christmas trees that will integrate selective pesticides and biological control.

Regional integrated pest management centers in the United States

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The United States Department of Agriculture's Cooperative States Research, Education and Extension Service funds four geographically-based Regional Integrated Pest Management (IPM) Centers (North Central, Northeastern, Southern and Western) to promote regional collaboration among universities in IPM research, education and regulation. Objectives of the Regional IPM Centers include: 1) organizing and maintaining multi-state IPM information networks; 2) involving stakeholders in identifying IPM needs and priorities within the regions; 3) managing regionally-based IPM grant programs; and 4) facilitating the development, review and communication of scientific and regulatory documents concerning IPM. The Southern Region IPM Center at North Carolina State University serves as a repository for IPM information from all four regions of the United States. More than 600 "crop profiles" (i.e., pest management profiles for U. S. agricultural crops generated by university scientists with funding from the Regional IPM Centers) are maintained in a searchable database on the World Wide Web (http://pestdata.ncsu.edu/cropprofiles/cropprofiles.cfm). Information on the Regional IPM Centers is available at the following website: http://www.ipmcenters.org.

Development and implementation of integrated pest and disease management (IPDM) in prune orchards in Australia

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Prunes (*Prunus domestica*) are grown in three areas in southern Australia. A study from 1999/2000 to 2002/2003 on 12 representative prune orchards in two areas in New South Wales identified the main pests as *Aculus fockeui* (Nalepa & Trouessart) (Acarina: Eriophyidae), *Tetranychus urticae* Koch (Acarina: Tetranychidae), *Quadraspidiotus perniciosis* (Comstock) (Hemiptera: Diaspididae) and *Maroga melanostigma* (Wallengren) (Lepidoptera: Oecophoridae). Main diseases were prune rust, caused by the fungus *Tranzschelia discolor* Tranzschel & Litv. and brown rot (*Monolinia* spp.). IPDM techniques were introduced to manage the pest and disease complex. Eight species of predatory mites (F. Phytoseiidae) were endemic in Murrumbidgee Irrigation Area orchards and two species in the Young area. Successful biological control of pest mites was demonstrated. Winter application of 2% dormant oil, with or without 1% horticultural mineral oil sprays timed to coincide with the first generation of crawlers was proposed as an IPDM alternative to the organophosphate insecticide methidathion for *Q. perniciosis*. Biological control of *M. melanostigma* is the subject of a separate study. Neither prune rust nor brown rot were serious problems during the investigation and were successfully managed with protective fungicides. Implementation of IPDM was promoted through workshops held in each of the three growing areas.

The suitability of different plants as food sources for larvae of garden weevil, and fuller's rose weevil.

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Garden and Fuller's rose weevil are pests of deciduous fruit orchards in Western Australia. Damage by these weevils includes poor orchard establishment, fruit downgrading, defoliation and blocking of mini-sprinklers. Methods of control are limited to the adults through the application of broad-spectrum insecticides. Disadvantages of which include inconsistent control, potential to cause secondary pest outbreaks and insecticide residues on fruit. A new research project examined the role of ground cover plants common in orchard interrows on the survival and growth of the soil dwelling larval stage. The aim was to identify suitable plants to reduce the abundance of the weevil larvae. This research was based on pot trials and single plant species. For garden weevil, survival on subterranean clover was very poor either from being an unsuitable food source or antibiotic activity. Subsequent research clarified clover as an unsuitable food source when grown with a commercial crop. The research protocol for assessing plants now includes a combination of a tree crop and a ground cover species.

Aphaereta debilitata **(Hym., Braconidae) as a biocontrol agent of the shore fly** *Scatella tenuicosta* **(Dipt.: Ephydridae)**

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The biological parameters of *Aphaereta debilitata* were determined and the parasitoid's efficacy tested in cages to assess the species' potential in controlling *Scatella tenuicosta*, a cosmetic pest of potted herbs, lettuce and seedlings in greenhouses. *Aphaereta debilitata* appears to be a solitary koinobiont endoparasitoid of *S. tenuicosta*. At constant 25^{o}C, females preferred host larvae of the age of 1 or 2 days to those that were <1 or 3 days of age at the time of parasitization. With continuous access to hosts, females lived 6.4 + 0.3 (s.e.) days, peaking their progeny production on day four at 43.5 daughters and sons. Life time progeny production was 215 (48% females). Males developed into adults in 13.0 ± 0.3 days, females in 14.0 ± 0.03 days. The intrinsic rate of increase of *A. debilitata* was 0.261 day^{-1}, the net reproductive rate R_0 was 85.3 female offspring per female, and generation time T was 5.6 days. In insect cages, the application of a total of 2.35 *A. debilitata* females per 8 x 8 cm peat pot at 4-6 days intervals resulted in control efficacy of 70-92 %, based on adult fly counts per pot every four days during the four week experiment.

The efficacy of three soil-living predatory mite species as predators of thrips pupae

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The predatory mites *Geolaelaps aculeifer* (a Queensland population GAQ, and a commercial population), *G. queenslandicus* and *Lasioseius* sp. were compared as predators of *Frankliniella occidentalis* pupae in peat pots using predator:prey ratios 2, 1, 0.5 and 0.25. The experiment was repeated three times. During 10 days, the commercial *G. aculeifer* had a control efficacy of 97.6% and 95,8% with predator:prey ratios of 2 and 1, respectively, followed by GAQ with efficacies of 91.2% and 88.6%. The control efficacy of *G. queenslandicus* was between 33.9% and 74.3% for all predator:prey ratios, and that of *Lasioseius* sp. between 41.5% and 70.9%. The predation rates of female GAQ and *G. aculeifer* were further compared on charcoal-Plaster of Paris arenas. During five days, GAQ-females killed 13,8 ± 3,14 pupae per day, and those of *G. aculeifer* 13,5 ± 3,22 (std) pupae per day. GAQ, however, laid statistically fewer eggs per day (4.4 ± 1.44) than *G. aculeifer* (5,0 ± 1,50). These short-term experiments with fixed prey densities need to be complemented with long-term experiments to see if differences in development times and reproduction rates of the mites are reflected in their long-term ability to suppress the prey in a manner different from the short-term experiments.

A rapid method in laboratory to screen resistance of potato plants against *Myzus persicae*

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In the objective to develop potato resistance against aphids, we propose a rapid screening method with *Myzus persicae* (Sulzer) in laboratory. Our study focussed on two objectives, i.e. to optimize the duration of the whole procedure and to decrease the frequency of measurements. In a first experiment, no significant differences were observed between intrinsic rate of natural increase (r_m) values of adult aphids reared throughout their entire life and adults reared only during a period equivalent to their pre-reproductive period. In a second experiment, four groups of aphids were distinguished according to the sampling frequency, i.e. those whose biological parameters were evaluated every single, second, third and fourth day. Except for the fourth day experiment, the r_m values estimated every single, second and third day were not significantly different for all potato lines. Thus screening efforts in laboratory can be largely optimized by checking adult aphids only during a period equivalent to their pre-reproductive period and assessing *M. persicae* populations every third day. Our method is reliable and adapted to screen a large number of potato plants against *M. persicae* as it leads to an average 70% reduction in the time required for the whole experimental process. Our laboratory screening method must be complemented with a field screening program.

Granulovirus for management of codling moth, *Cydia pomonella* L. (Tortricidae)

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Codling moth granulovirus -based insecticides were assessed in apple orchards in Washington State, USA, British Columbia and Quebec, Canada, against *Cydia pomonella* L. (Tortricidae). In all studies, the virus provided effective control of neonate codling moth. CydX, Virosoft CP4, and Carpovirusine were assessed in orchard trials. All treatments remained significantly effective after 14 days. When Virosoft CP4 was applied alone and with Surround (kaolin), seven sprays of Virosoft CP4 provided adequate codling moth control and Surround did not increase the pathogenicity of CP4. In semi-field trials, significantly higher codling moth mortality was recorded after neonates fed on apples from trees treated with the virus compared to untreated apples up to one and eight days post-treatment with Carpovirusine and Virosoft CP4, respectfully.

IPM of *Xyleborus fornicatus* Eichh. (Coleoptera: Scolytidae), the shot-hole borer of tea in Sri Lanka

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The shot-hole borer first recorded in 1892 is one of the most destructive pests of tea grown in Sri Lanka at altitudes between 150m to 1350m. The concealed habit of this pest makes it notoriously difficult to control. The pesticide approach using persistent organochlorine, DDT in 1940 and its successors like endrin, aldrin and dieldrin, lead to secondary outbreaks of other minor pests like Twig & looper caterpillars. This led to the development of the concept of IPM in tea production in parallel to the trend all over the world in 1970's. The elements of IPM include the use of varietal resistance, an escape strategy where pruning is done to avoid the overlap of the susceptible primary branch stage of the plant with peak colonization period of the beetle, sanitation pruning to reduce the wood rot in subsequent cycles with limited use of fenthion (Lebycide®), an organophosphate insecticide currently in recommendation. Biological control has not been successfully employed against the pest but recently there were some indications that the fungus *Beauvaria bassaiana* could be used effectively in controlling the beetle and hence, another prospective tactic for the IPM programme in the future.

Morphological and cytological changes in leaves of cucumber and tomato injured by western thrips, *Frankliniella occidentalis* Pergande

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Direct damages due to feeding of western thrips on plant's juice result from injuring the epidermis and mesophyll. Morphological changes of the epidermis of cucumber leaves are typified by the presence of amorphous structures generated on the surface of the cuticle. The stomatal apparatuses become invisible and the hairs on parts of the disturbed epidermis are subjected to deformation. On under surface of the epidermis instead, a collapse of tissue is observed. Formation of amorphous structures is here present too, but unlike the upper surface, the stomatal apparatuses are opened. Amorphous changes on a tomato leaf injuried by western thrips are decidedly more soft as compared to cucumber, however, the hairs within the reach of the injury are more deformed. Cytological changes in cucumber are visible as necroses. They are generated due to injuring the cells of both the upper epidermis and palisade mesophyll. Extremely, the complex of deformed cells expands from the upper epidermis beginning, through palisade and spongy mesophyll up to the under epidermis. In tomato, at first inconsiderable necroses within the epidermis appear. They can reach as far as palisade or even spongy mesophyll, but allways they are smaller in size, as compared to cucumber.

A systems approach to IPM for organic apple production in New Zealand

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Paper not available at time of production.

IPM of protected cropping in developing greenhouse regions.

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Paper not available at time of production.

Modelling winter flight activity of sterile Queensland fruit fly, Bactrocera tryoni (Frogatt) (Diptera: Tephritidae) in Leeton, Australia

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Sterile Queensland fruit fly (Qfly), Bactrocera tryoni (Diptera: Tephritidae), were released across the township of Leeton, New South Wales, Australia, as part of an eradication program. Releases were conducted until mid May 2003 and targeted urban households and adjoining orchards. Although releases did not resume until mid August 2003, sterile males were caught in cue-lure baited Lynfield traps throughout June, July and August. Flies released in autumn are generally able to achieve sexual maturity and remain attracted to cue-lure. Trap records suggest that flight is possible at certain times during winter. This paper examines climatic data for the period June through August 2003 for the Leeton area in relation to modeling flight activity of Queensland fruit fly. Hourly data was corrected for daylight hours, rainfall, wind-chill and tree-canopy wind-drag variables for the period. When corrected only for daylight there was found to be a total of 192 hrs where flight was possible. This was reduced to 136 hrs when further corrected for wind-chill, and to 94hrs including wind-drag corrections. Rainfall events were few and reduced these figures to 179 hrs, 129 hrs and 82 hrs respectively. Wind-chill had the most influence on reducing potential flight periods. These results are discussed in relation to other data formats and behavioural aspects of Queensland fruit fly.

Developing infochemical-based tools and management methods for giant wood moths (Endoxyla cinerea) and longicorn beetles (Phoracantha spp.) in Queensland

SA Lawson

Paper not available at time of production

Dispersal and host location by Lygus spp.: Underlying behavioral and ecological mechanisms and implications for IPM

JL Blackmer

Paper not available at time of production

IPM programs as influenced by market destination

K Heinz

Paper not available at time of production

Effect of Body Size on Reproductive Fitness in Aphidius ervi (Hymenoptera: Aphidiidae)

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Aphidius ervi Haliday is an important parasitoid of several aphid species on economically important crops such as legumes and cereals. To improve its mass rearing and field release efficiency, we studied the relationships between body size and reproductive fitness of A. ervi on pea aphid, Acyrthosiphon pisum (Harris), in the laboratory. Large females lived significantly longer (14.10 ± 0.72 d), had significantly higher searching efficiency (1.50 ± 0.37 10^{-4}min), fecundity (574.25 ± 23.76 eggs) and fertility (338.74 ± 19.41 eggs), and parasitized significantly more aphids (410.70 ± 17.74) than did small females (11.60 ± 0.57 d, 0.97 ± 0.09 10^{-4}min, 302.74 ± 17.61 eggs, 213.39 ± 15.17 eggs and 257.07 ± 14.18 aphids for longevity, searching efficiency, fecundity, fertility and parasitism, respectively). However, fertility rate was significantly higher for small females (0.6923 ± 0.0238) than large females (0.5819 ± 0.0252). Male size was positively correlated with fertility ranging from 246.60 ± 20.43 eggs for small males to 305.53 ± 20.02 eggs for large males, and with fertility rate ranging from 0.5771 ± 0.0276 for small males to 0.6971 ± 0.0199 for large males. Male size had no effect on fecundity and parasitism. Implications from this study were discussed.

Bee-eaters: Ravager of beekeepers but saver of farmers

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The wild birds have been offered food and water as a traditional custom in many religious communities in Asia for centuries as well as Australian feed their domestic birds. Just as the expensive name reveals, bee-eaters predominantly eat huge number of insect pests and other invertebrate pests. All insectivorous birds can eat harmful herbivorous insects to keep under control of pest populations in agro ecosystems. Bee-eaters consume a large number of bees causing direct impact on quantity honey production during honey flow season in Thailand (Singh, 1962; Jenn, 1973; Esmaili, 1974; Buys, 1975; Fry, 1984). Mostly they become to be the honeybee pest when *Apis mellifera* was introduced to the country and during the migration seasons of native bees (*A. cerana*, *A. dorsata* and *A. florea*).

Key arthropods for the control of *Epiphyas postvittana* (Walker) (Lepidoptera: Tortricidae) in Coonawarra vineyards, South Australia

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Epiphyas postvittana (Walker) light brown apple moth (LBAM) is indigenous to Australia and a major pest of horticultural crops including grapevines. We conducted studies to identify predatory and parasitic arthropods of LBAM, from Coonawarra vineyards in South Australia and investigate the density-dependant response of key parasitoids to varying host densities of LBAM in the field. Arthropods in Coonawarra vineyards were surveyed for 12 consecutive months using pit-, pan-, sticky-traps and beating techniques. More than 60 species of beneficial arthropods were identified. However few occur in the canopy. The species that attack LBAM include a predatory mite *Anystis baccarum* and four parasitoids. The most dominant parasitoid is a braconid *Dolichogenidea tasmanica* (Cameron) followed by two species of Ichneumonidae and a tachinid *Voriella uniseta*. To evaluate the response of hymenopteran parasitoids to varying host densities we experimentally manipulated the number of larvae in vineyards on three occasions. Our results show that parasitism at the scale of a panel (four grape vines) was inversely density-dependent. Percentage parasitism was 29% and 38% for high and low host densities respectively. Evaluating predation and parasitism will contribute to moving one step closer to integrated pest management of LBAM in Coonawarra vineyards.

Citrus pests and their natural enemies; integrated pest management in Australia – a photographic tour

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The book 'Citrus pests and their natural enemies; integrated pest management in Australia', and its companion, 'Citrus pests a field guide' were published in 1997. The CD (an updated, html version) 'The Good, the Bug & the Ugly', was published in 2003. These publications represent the state of the art in Australian citrus IPM, distilled from the experience of numerous researchers, field scouts & extension officers. Reviews, local and overseas, have been very favourable, these volumes regarded as exemplifying a highly readable and practical approach to the implementation of IPM. Invariably, reviewers have complemented the quality and variety of the more than 400 colour photographs, many of which show close-ups of live insects in action. In this paper a high impact selection of these photographs, most of which were taken by its authors, will be projected, organised along the same lines as the book. That is - Mites, Ants, Soft scales, Hard scales, Mealybugs, Aphids, Planthoppers & leafhoppers, Cicadas, Whiteflies, True bugs, Moths & Butterflies, Beetles, Katydids crickets & grasshoppers, Flies & midges, Wasps, Thrips, Termites and Spiders. Special emphasis will be given to the beneficials - parasitoids & predators - crucial to the success of citrus IPM in Australia.

Behavioural fever in acridids: potential impact on biological control

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Arthropods can respond to microbial infection by developing a behavioural fever. We assessed the occurrence of behavioural fever in seven acridid species from different ecological habitats, using three entomopathogenic fungi. Except for one species, all infected insects developed a behavioural fever as their preferred body temperatures were usually higher than those of healthy, uninfected counterparts. The three pathogen species elicited behavioral fever, athough differences were observed among fungus-insect associations. We used the association between *Locusta migratoria* and *Metarhizium anisopliae* to further study the implication of such behaviour on both the host and the pathogen. Variations of thermoregulation regimes affected insect mortality: mortality decreased with increasing duration of thermoregulation, delayed onset of thermoregulation increased mortality, and interruption of thermoregulation was followed by resumption of mycosis. In the absence of thermoregulation, hemocyte concentrations decreased dramatically, along with a colonization of hemolymph by blastospores. The phagocytic activity was hampered in locusts that were not allowed to thermoregulate. We further demonstrated a differential protein expression between thermoregulating and non-thermoregulating locusts. These results indicate that infection-induced behavioral fever is common in acridids and may limit the potential of fungal pathogens as biological control agent under certain ecological conditions.

Release of *Peristenus digoneutis* (Hymenoptera: Braconidae) in Canada for biological control of *Lygus lineolaris* (Hemiptera: Miridae)

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This national project targets suppression of *Lygus* spp. in non-cropping and low-intensity situations by releases of the European parasitoid *Peristenus digoneutis* [Hymenoptera: Braconidae], thereby decreasing pest levels in adjacent agricultural crops. Pre-release parasitism by native larval parasitoids, *Leiophron* and *Peristenus* spp. [Hymenoptera: Braconidae], on *Lygus lineolaris* in Ontario, was consistently below 10%. From 2001 to 2003, more than 11,000 *P. digoneutis* from Europe were released on alfalfa in London, Ontario. We monitored parasitoid overwintering, and dispersal and potential non-target hosts. In the lab we have investigated interspecific competition of parasitoids; the acceptance and suitability of non-target mirid hosts including the predatory mirid *Dicyphus hesperus*; and interaction of two biological control agents: an imported parasitoid and the entomopathogenic fungus *Beauveria bassiana.*

Eretmocerus mundus Failure to Control *Bemisia tabaci* on Sage

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Bemisia tabaci (Grennadius) is a serious polyphagous pest of many agronomic and ornamental crops on all continents. Of the herbs grown for fresh-market in Israel, sage (*Salvia officinalis* L.) is a particularly problematic crop due to its susceptibility to whitefly infestations. Studies were conducted to determine the effect of the parasitoid, *Eretmocerus mundus* Mercet (Hymenoptera: Aphelinidae) on *B. tabaci* infesting sage grown in walk-in tunnels. *Eretmocerus mundus* pupae were obtained from a commercial insectary and released at total rates of 10, 20 and 30/m^2. No *E. mundus* parasitized nymphs were found; however, there was natural parasitization by *Encarsia* spp. In contrast, parasitization by *E. mundus* released in neighboring basil tunnels was about 5 – 16%. Laboratory trials to confirm these findings on sage were conducted. Since *E. mundus* lays its eggs under the whitefly nymph, it is suggested that the morphological changes in the nymphs due to the densely hairy leaf surface are responsible for these results.

New opportunities for biological control of olive fruit fly

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Bactrocera oleae is a key pest of olives in the Mediterranean region and in California. Indigenous natural enemies do not provide adequate control in Europe, and early attempts to introduce parasitoids from South Africa were unsuccessful. Although the fly is often assumed to be native to the Mediterranean region its origin remains an open question. It occurs widely in Africa and Asia where cultivated and wild olives are found but little is known regarding its biology and ecology in wild olives. Previous surveys in Africa early in the 20th century revealed a rich diversity of olive fly parasitoids not present in the Mediterranean basin. Other parts of the fly's range have never been surveyed for natural enemies and it is likely that additional diversity in natural enemies remains to be discovered. Exploration for natural enemies offers an excellent opportunity to examine the genetic variability in populations of the fly, which may help to answer questions regarding the native range of the olive fly. Renewed foreign exploration throughout the range of olive fly was initiated in 2000. African and Asian collections to date have yielded new information on the ecology of olive fly in wild olives, which will be reviewed. Entomopathogens may also have a role in olive fly management.

Importation, host specificity testing, establishment and assessment of *Diversinervus* sp. near *stramineus* Compere (Hymenoptera: Encyrtidae), Kenyan parasitoid introduced for green coffee scale Coccus viridis (Green) in Queensland and northern New South Wales.

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The Encyrtid parasitoid *Diversinervus* sp. near *stramineus* Compere, was introduced from coastal Kenya to Australia for the biological control of green coffee scale, *Coccus viridis* Green, infesting citrus, coffee and ornamentals. After quarantine testing against 16 other native and introduced insect scales and mealybugs, *D.*nr.*stramineus* was found to be specific to *C. viridis,* and releases were made throughout Queensland and northern New South Wales. The parasitoid's efficacy was assessed in three citrus blocks in Southeast Queensland and in a coffee block in North Queensland over three-four years. The parasitoid established readily, becoming the dominant natural enemy within 12 months of release, with parasitism levels of up to 80%. *D.*nr.*stramineus* has established throughout Queensland and northern New South Wales, and together with the existing Aphelinid parasitoids, *Coccophagus ceroplastae* (Howard) and *Eurischomyia flavithorax* (Girault and Dodd), and the fungal pathogen, *Verticillium lecanii* (Zimmerman), has reduced *C. viridis* to minor pest status.

Three other parasitoids *Metaphycus baruensis* (Noyes), *Metaphycus stanleyi* Compere and *Coccophagus bogorensis* (Koningsberger), were introduced to quarantine but not successfully reared on *C. viridis*.

Surveillance and control of exotic mosquitoes in metropolitan France

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Aware of the importation risks of exotic mosquitoes into West European countries, an entomological surveillance has been set up in France. The main risk is the introduction and dissemination of the tiger mosquito *Aedes albopictus* by the used tyre trade. Investigations at the main used tyres importers' sites have permitted the detection of *Ae. albopictus* in France in 1999, then in Belgium in 2000. A second Asian mosquito, *Ochlerotatus japonicus*, as well as a North-American mosquito *Oc. atropalpus* have also been discovered in 2000 and 2003 respectively. *Aedes albopictus* and *Oc. japonicus* are potential vectors of arboviruses (such as West Nile virus) and dirofilariasis. Moreover, *Ae. albopictus* is responsible of an important biting nuisance in all the colonised temperate regions. Aware of this, the French public mosquito control agencies decided in 2001 to stop the expansion of the species and therefore to treat the infested sites. Surveillance and control operations are supported by the French Ministry of Health. Due to the permanent reintroduction risks, the surveillance is extended and an immediate spraying is applied on all the colonised sites. Regular surveys and rapid action should, if not stop, at least delay the settlement in France of exotic potential vectors mosquitoes.

Fungal biopesticides for control of aphids in cotton and grains.

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Microbial insecticides have been shown to be a valuable component of integrated pest management in Australia for control of *Helicoverpa* sp. in broadacre crops. We are evaluating potential new biopesticides based on entomopathogenic fungi for control of aphids in cotton, sorghum and pulses. We have assessed 50 isolates of *Metarhizium* and *Beauveria* for activity against the cotton aphid (*Aphis gossypii).* Laboratory assays have shown three *M. anisopliae* isolates and one *B. bassiana* isolate have significant potency against aphid nymphs at five days post infection. A field trial in cotton compared efficacy of *Metarhizium* isolates, and showed that one isolate significantly reduced aphid populations compared to unsprayed controls. Further work will focus on improving the efficacy of the agents through formulation and targeting of pests.

The biological control of Kelly's citrus thrips in Australian citrus orchards.

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Kelly's citrus thrips (KCT) (*Pezothrips kellyanus* (Bagnall)) (Thysanoptera: Thripidae) has emerged in the 1990's as a serious citrus pest in southern Australia, New Zealand and parts of the Mediterranean Basin. The feeding of KCT on young and mature fruit causes scurfing (or halo) marking and rind bleaching, respectively. These blemishes reduce fruit quality, thereby reducing the packout of export quality fruit and rendering some fruit unsaleable. Information about KCT biology and ecology has been scarce, and in Australia the management of KCT has been limited to the use of several insecticides. Having established that KCT pupate in the soil below the citrus canopy, we initiated a search for both arboreal and soil-dwelling natural enemies with the aim of developing an IPM system for KCT in Australian citrus orchards. The study has identified a range of mite species that are either demonstrated or likely predators of KCT. The relationship between the diversity and abundance of these predators and the incidence of KCT in study orchards is examined. The potential implications for the IPM of KCT are discussed.

Sirex wood wasp oviposition in *Pinus caribaea* & *P. caribaea x P.elliottii* hybrids.

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Sirex wood wasp (*Sirex noctilio*) is a destructive introduced pest of *Pinus radiata*, which is the main exotic softwood plantation species in New South Wales, A.C.T., Victoria, South Australia and Tasmania. Sirex was detected in mainland Australia in 1962, and posed a major threat to the softwood industry. The successful implementation of a biological control program reduced the population to low levels but it spread, and is now close to the Queensland border. In Queensland the exotic softwood plantation estate is mainly *Pinus caribaea, P. elliottii* and *P.caribaea x P. elliottii* hybrids. The potential of *P. caribaea* and the hybrids to host Sirex or its biological control agents was unknown and the threat to Queensland plantations could not be assessed. We investigated the susceptibility of *P. caribaea* and the hybrids to Sirex infestation. Billets of *P. caribaea* were exposed to field-collected *P. radiata* billets with newly emerging Sirex and parasitoids. Billets of *P. taeda* and the *P. caribaea x P. elliottii* hybrids were exposed to female Sirex wasps newly emerged from field-collected *P. radiata* billets. During and following exposure the billets were stored in an insectary. We comment on susceptibility and development times.

Video monitoring nocturnal predation of *Helicoverpa armigera* larvae

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Numerous studies have looked at diurnal predators of *Helicoverpa armigera* but few researchers have attempted to study nocturnal predation of immature and adult *H. armigera.* In our current study we are using 4 digital B&W cameras fitted with infrared illumination. Each camera records the nightly activity in a 50 x 50 cm arena of soil surface in the inter-row space. A four-channel digital recorder stores images at 1 frame per second on a removable 80 GB hard disk drive. Trials have been conducted in summer field crops including maize, cotton, mungbean and sorghum. For each arena we tethered sentinel prey (5 helicoverpa larvae or moths) and recorded which predators consumed them. We recorded the activity of predators over the duration of the night and the time taken to consume all prey. Pitfall traps were used to aid identification of predators active at each site. A species list of animals recorded by the cameras has been compiled for each site and includes field mice (*Mus musculus),* carabid beetles (*Gnathaphanus pulcher* and, *Helluo costatus),* predatory earwigs (*Labidura truncata)* and wolf spiders *(Lycosa godeffroyi).* These video studies identified several important predators that have potential to reduce the numbers of large larvae surviving through to pupation.

Effect of transgenic potato (GNA) in the tritrophic system with aphids (*Myzus persicae*) and aphid parasitoids (*Aphidius colemani*).

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A gene encoding snowdrop lectin (*Galanthus nivalis* agglutinin) has been engineered into a number of crops, including potato. Experiments have shown that such GNA potato plants are partial resistant towards aphids, and have a potential to affect the aphid parasitoids, which are important biological control agents. We have studied effects of GNA potato on the aphid *Myzus persicae,* and the aphid parasitoid *Aphidius colemani.* No significant effects have been found on aphid development or fecundity. Preliminary results on the searching behaviour, development time, longevity and adult size of *A. colemani* are presented. All experiments have been conducted in laboratories using transgenic plants, instead of artificial diet, as the transformation process can cause unexpected effects that might influence the insects.

Assessment of *Trichogramma embryophagum* **(Hymenoptera:Trichogrammatidae) against** *Sesamia* **(Lepidoptea:Noctuidae)**

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The role of *Trichogramma embryphagum* (Hymenoptera:Trichogrammatidae) (reared on *Heliothis zea* Boddie eggs) as parasitoid of the sugarcane and corn stem borers *Sesamia cretica* Led. And *S. nonagrioides botonephaga* T.&B.,was assessed under laboratory and greenhouse condition .Data on seven biological parameters are presented.The laboratory results showed that *T.embroypham* reared on *H. zea* eggs parasitized 50% of *S.critica* gggs,compared to 58.5% of *S .nonagrioides* eggs.percentages of emergence (87%), female progeny (71%),deform females (2.5%), and development time (8.9d) in *T. embroypham* reared from *S. cretica* gggs did not differ significantly from those reared on *H. zea* eggs (93%,69%,3.2%,2d and 8.5d respectively).However,*T. embroypham* females reared form *S. cretica* eggs had greater body length (39mm) than females reared on *H. zea* (0.33mm). in the greenhouse conditions, *T. embroypham* parasitized an average of 56.4% of the *S.cretica* eggs masses were distributed on the corn plants).about 21.6% of the eggs were dessicated,probably due to puncturing by the parasitoid,compared with 4.1% in the control,where egg masses were distributed on the plants without release of *Trichogramma*. Results showed the potential for use *Trichgramma* to control corn stalk borers in a field IPM program.

Historical perspective on *Xenorhabdus*/ *Photorhabdus* research

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Xenorhabdus and *Photorhabdus* are Gram negative bacteria that are symbiotically associated with entomopathogenic nematodes of the families Steinernematidae and Heterorhabditidae, respectively. They have progressed from being a microscopic oddity, through being recognised as critically important for the success of their nematode partners to being a key element of a range of biological insecticides. Along the way, they have raised some intriguing scientific questions and developed as sources of insecticidal and antibiotic activity in their own right. The discovery of a *Photorhabdus* sp. in human clinical specimens opened a new view on their evolutionary history. The recent sequencing of a *Photorhabdus* genome increases the attractiveness of these bacteria as a model system for the investigation of symbiotic relationships and of pathogenesis.

Rearing the potential predator *Geocoris lubra* Kirkaldy (Hemiptera: Geocoridae).

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Geocoris lubra Kirkaldy (Hemiptera: Geocoridae) or the big-eyed bug, is considered to be a major predator of *Helicoverpa armigera* Hübner (Lepidoptera: Noctuidae) in Australian crops – particularly cotton and soybean. *G. lubra* is therefore of potential value as a biological control agent of *H. armigera* in pest management strategies such as integrated pest management. Despite this, very little is known about the biology and ecology of *G. lubra* and until recently a rearing method had not been developed. We describe a successful rearing method for *G. lubra* and determine the impact of photoperiod on aspects of *G. lubra*'s biology. Temperature was found to be the key parameter required for successful rearing of the predatory bugs. However, photoperiod could greatly increase the percentage of viable eggs and significantly shorten development time from egg to adult.

"Integration of transgenic rice with IPM"

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"Transgenic rice lines with *Bt* and *Gna* genes against stem borers and hopper pests, respectively, are being field evaluated in India. While greenhouse and initial limited field tests have identified some of the lines to be promising against the target pests, no extensive data on field performance of these lines, impact on non-target arthropods are available yet. However, based on our experience of rice insect pest management in India we anticipate following opportunities, problems and challenges ahead in deploying transgenic rice as component of rice IPM. It is imperative that transgenic rice line proposed to be deployed needs to be effective not only against most species of the target stem borers and hoppers but also against non target pests like leaf folders, if farmers are to refrain from insecticide application. Since natural biological control is effective under most circumstances, study on effect of transgenic line on natural enemies is critical. Distinct economic impact of transgenic rice will be felt only in some parts of the country and hence may act as disincentive to farmers in other parts if seed cost and grain quality does not match their expectation. There may not be major barriers to integrate transgenic rice with IPM.
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Dispersal, biology and impact of the introduced biocontrol agent *Zygogramma bicolorata* Pallister (Coleoptera : Chrysomelidae) on the weed *Parthenium hysterophorus* L. in Jammu (J&K), India

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Parthenium hysterophorus L. is a weed of national significance in India. It is the most predominant weed of wastelands, pastures and agricultureal fields and has spread throughout India. It has achieved the status of a worst weed owing to its adverse effects on human beings, animals, and natural diversity. In Jammu and Kashmir state, parthenium has spread to all the six districts of Jammu division and is moving fast and far in other high altitude areas including the Kashmir valley. Biological control trials against this weed were initiated in Jammu region by the release of the leaf - feeding beetle *Zygogramma bicolorata.* This beetle established readily and started building up damaging population levels and has dispersed to most parts of Jammu causing large scale defoliation of parthenium. In this study we have recorded the dispersal of this beetle in Jammu and studied its biology and performance in different climatic conditions. Besides, impact of defoliation by *Z .bicolorata* on parthenium has been discussed.. The results of the studies carried out to date indicate that *Z .bicolorata* has the potential to bring about reduction in the density of parthenium in Jammu.

Functional morphology of parasitoid immatures

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The immatures of egg parasitoids develop inside host eggs and, because of their minute size and their special habitat, are considered as morphologically specialized when compared to other Hymenoptera immatures. Among the constraints of using insect eggs as host, the small size of insect eggs and the short period during which the host stage is available are major forces that shaped the behavior and physiology of egg parasitoids. Most egg parasitoids are idiobiont species and their immatures have to develop on the finite resources within the host egg. These immatures may either share these resources with conspecifics, in the case of gregarious species, or eliminate surnumerary conspecifics in the case of solitary species. Both physiological and morphological adaptations to this competition are found, mostly in the first instar larvae. The host egg is more than a simple container for the parasitoid immature and its chorion is important in the regulation of oxygen and water within the egg. In some egg parasitoid species, successful development is only possible in host eggs where the chorion has completed its formation while some other species perforate the chorion to access directly atmospheric air.

Recombinant baculovirus insecticide expressing a novel intrahemocoelic toxin

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Basement membranes surrounding the tissues of lepidopteran larvae serve as a potential barrier to baculovirus movement and establishment of systemic infection. To counteract this barrier, we constructed a recombinant *Autographa californica* nucleopolyheovirus (AcMNPV) that expressed a basement membrane-degrading cathepsin L protease from the flesh fly, *Sarcophaga peregrina*. In survival time bioassays, this recombinant, AcMLF9.ScathL, killed neonate *Heliothis virescens* 50% faster than wild-type virus and 30% faster than viruses expressing the scorpion toxins AaIT and LqhIT2 (AcMLF9.AaIT, AcMLF9.LqhIT2). Baculovirus-expressed *S. peregrina* cathepsin L was optimally active at pH 5.0 and not active at pH 6.0, suggesting that *S. peregrina* cathepsin L may be operating at locally acidified microenvironments within the larvae. AcMLF9.ScathL caused premature cuticular melanization and melanization of internal tissues. Infections of fifth instar *H. virescens* with AcMLF9.ScathL yielded fewer polyhea than wild-type AcMNPV and AcMLF9.AaIT, indicating that the insecticidal properties of *S. peregrina* cathepsin L may be independent of any effect on virus movement or systemic infection. Risk assessment studies indicated that AcMLF9.ScathL had no detrimental impact on two nontarget insect predators, the ladybeetle *Coleomegilla maculata* and the green lacewing *Chrysoperla carnea*.

A mechanistic and predictive framework for predicting the efficacy of biocontrol agents: the application of competition and keystone predation theory to invasive species.

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We tested a rigorous approach for predicting whether insect herbivores will be effective biocontrol agents. Resource competition theory predicts that dominant plant species (whether exotic or not) are capable of persisting at the lowest level of a shared limiting resource or reducing the shared limiting resource more than competing species. Furthermore, keystone predation theory predicts that herbivores will alter competitive outcomes only if they sufficiently impede competing species from acquiring or reducing the limiting resource. We field-tested these two theories for 12 plant species (invasive and native) in both terrestrial and wetland habitats. Though work in the terrestrial habitat remains preliminary, in wetlands, our results suggest that the release of a leaf-feeding Chrysomelid beetle (*Galerucella calmariensis*) will not halt the spread and dominance of the invasive plant purple loosestrife (*Lythrum salicaria*) in the USA. Nonetheless, an enormous amount of resources have already been invested into releasing this beetle throughout the USA.

In vitro antimicrobial effects of herbal extracts on pathogens causing chalkbrood and European foulbrood diseases in honey bee (*Apis mellifera*)

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Brood diseases in honeybee (*Apis mellifera*) found in Northern Thailand during 2001-2002 were Chalkbrood and European foulbrood. Based on their characteristic symptoms, European foulbrood pathological samples were collected from Chiang Mai, Chiang Rai, Payao, and Phrae Province whereas Chalkbrood samples were collected from six apiaries (five apiaries in Chiang Mai, and one in Payao Province). Six isolates grown in anaerobic condition were *Melissococcus plutonius*, an European foulbrood pathogen. Endospore forming bacilli found in culture were identified as *B. stearothermophilus*, *B. macerans*, *B. laterosporus*, *B. circulans*, *B. firmis*, *B. coagulans*, and *B. licheniformis*. None of them was *Paenibacillus larvae* subsp. *larvae*, a causative agent causing American foulbrood. Forty-six isolates of fungi were identified as a Chalkbrood pathogen (*Ascosphaera apis*). Our aim is to control the diseases by using natural products, the screening of potential compounds from medicinal plants, therefore, was carried out. Dichloromethan extract of *Glycyrrhiza glabra* exhibited the best inhibitory effect on all *M. plutonius* strains (both Thai and Australian strains). Hexane *Cinnamomum cassia* extract and Ethyl acetate *Piper betle* extract gave good inhibitory effects on *A. apis* growth. The cocktail of these selected herbs is proposed as an alternative for potential disease control.

The biological control programme for the obscure mealybug, *Pseudococcus viburni*, **using** *Pseudaphycus maculipennis* **(Hymenoptera: Encyrtidae)**

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Pseudaphycus maculipennis is one of the few known encyrtid parasitoids of *Pseudococcus viburni*, and was recognised as a potential and practical biocontrol agent for this mealybug in New Zealand in the early 1990s. *P. maculipennis* was imported into quarantine in Auckland in December 1997, before new environmental legislation (the Hazardous Substances and New Organisms Act) was implemented. After host testing in quarantine, application for approval to release was made to the Environmental Risk Management Authority in April 1999. Approval to release was given on 9 August 2000, after extensive consultation and provision of further technical information, and a public hearing. The first *P. maculipennis* were released in February 2001. By 2004, approximately 765,000 *P. maculipennis* ("Peemac") had been released to nearly 50 pipfruit orchards in Hawke's Bay, Nelson/Motueka and Auckland, and to the Wellington Botanic Gardens. *P. maculipennis* has dispersed at least locally, but no studies of its impact on obscure mealybug populations have yet been carried out. Using 'trap' mealybugs, Peemac have been recovered from c.50% of the release properties over 2-3 years, and it is concluded that the species has established in New Zealand.

Identification, synthesis and exploitation of the pheromone for the goldeneyed lacewing, *Chrysopa oculata*.

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Four distereomers of iridodial have been synthesized in 5 steps from 4a*S*,7*S*,7a*R* and 4a*S*,7*S*,7a*R*-nepetalactones, major components of catnip oil. In the field-trapping experiments, 1*R*,2*S*,5*S*,8*R*-iridodial was the only male-specific compound that exhibited significant attraction to *Co. oculata* males. 1*R*,2*S*,5*R*,8*R*-Iridodial is identified as a male-produced male-aggregation pheromone for *Chrysopa oculata*, the first pheromone of any kind for lacewings. Preliminary studies using 1*R*,2S,5R,8R-iridodial for the practical utility in manipulating natural or artificially augmented populations of lacewings for enhanced biological control will be discussed.

Recruitment of egg parasitoids by oviposition-induced synomones: a review.

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"Studies that have investigated herbivore-induced volatiles have focused primarily on insects that damage plants by chewing. However, plants also respond to herbivore egg deposition by releasing volatiles which can attract egg parasitoids. Plants that are able to attract egg parasitoids soon after herbivore eggs are laid benefit from increasing the parasitoid's effectiveness and from shielding themselves against insect pests before any damage can occur. Evidence on oviposition-induced synomones has been reported for only three tritrophic systems. In two of these, *Ulmus minor* and *Pinus sylvestris* respond to oviposition of their herbivores, *Xanthogaleruca luteola* and *Diprion pini* respectively, by emitting volatiles that attract the *Oomyzus gallerucae* and *Chrysonotomyia ruforum*. The third tritrophic system consists of *Vicia faba* and *Phaseolus vulgaris*, which, under the combined feeding and oviposition activity by a piercing/sucking herbivore, *Nezara viridula*, emit volatiles that attract *Trissolcus basalis*. We will present data on the attraction of the egg parasitoid by oviposition-induced plant volatiles, the mechanisms of induction by egg deposition, and the specificity and the chemistry of volatiles induced, showing similarities and differences between the three tritrophic systems.
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Potential and risks of deploying transgenic entomopathogens

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Insect pathogens have shown considerable potential as control agents of insect pests, however their widescale adoption has been limited by concerns about factors such as their relatively slow speed of action, narrow host range and variable environmental persistence. Genetic manipulation has been proposed as one route to circumvent these limitations. The first wave of transgenic entomopathogens mainly addressed the issue of speed of kill as this trait has a direct impact on crop damage. In insect baculoviruses in particular, numerous laboratory studies have shown that the insertion of foreign genes can result in a fast-acting pathogen. More importantly, field trials also indicate that these changes can result in reduced crop damage. While it is clear that genetic manipulation can produce entomopathogens with altered characteristics likely to enhance their potential as pest control agents, these transgenic organisms cannot undergo widescale release into the environment without appropriate ecological risk assessment. The limited risk assessment studies carried out to date indicate that the introduction of foreign genes is unlikely to result in a pathogen with enhanced fitness. However further studies are needed, in particular relating to longer term persistence, alternative hosts and mixed pathogen infections.

Factors defining *Trichogramma* abundance in the Ord River Irrigation Area in remote northern Australia

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Advances in insect pest control technology and renewed interest in re-establishing a cotton industry in tropical northern Australia led to recent cotton trials in the Ord River Irrigation Area (ORIA). The introduction of transgenic cotton varieties and modern Insect Resistance Management (IRM) techniques effectively minimized chemical pesticide use. Eggs of *Helicoverpa* collected during trials produced high numbers of *Trichogramma* egg parasitoids. Their impact on the potentially resistant pest species *Helicoverpa armigera* (Hübner) is considered integral to the IRM strategy for transgenic cotton production in the ORIA. Our research examined aspects of *Trichogramma* ecology pertinent to this strategy. Principal investigations focused on host egg collection providing indications of *Trichogramma* activity. Measured as percentage parasitism, *Trichogramma* activity appears highly variable in space and time and does not necessarily coincide with periods of peak insect pest abundance. Host abundance alone does not define conditions suitable for *Trichogramma* activity. Environmental constraints on wasp survival, such as the impact of temperature, humidity and chemical applications, are equally important factors limiting their effectiveness in biological control. Subsequent investigations targeted survival and distribution of adult *Trichogramma* to complement host interaction data. The aim was to isolate factors that limit the biological control potential of *Trichogramma* in the ORIA.

Present status of olive fruit fly SIT development

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The olive fly is the sole key insect pest for the olive tree in most of cultivated olive tree areas around the world. Among others, extensive research has been undertaken on the sterile insect technique against the olive fly. In spite of more than 20 years of research and development (1963-1983), no effective application of the methodology has been achieved. Consequently, since about 1980 the emphasis on this particular methodology with the olive fly has progressively diminished. The main obstacles connected with the difficulties of SIT with this particular insect were: laborious and expensive mass rearing, reduced quality of mass reared flies (e.g. different mating time than the wild insects under natural photoperiod), and lack of essential information on the complete role of fly's symbionts, and population changes in nature. Recently, work has been resumed in 3 main directions: a) developing cheaper, less "deleterious" to the fly (because of antibiotic, preservatives) and of more practical use diets, b) exploring in detail and possibly resolving the problem of differential mating time of laboratory insects as compared to wild ones, and c) developing transgenic strain(s) towards practical mass separation of sexes.

Biological control of pest mole crickets in Florida, USA

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Three South American pest mole crickets occur in Florida. They arrived in Georgia or Florida in 1899-1904 in ship ballasts. For >100 years, chemicals were the control method, at great annual cost, in vegetables, turf, and pastures. In the 1980s, three specialist biological control agents were introduced into northern Florida against them, all from southern South America. These were a wasp, *Larra bicolor* (Hymenoptera: Sphecidae), a nematode, *Steinernema scapterisci* (Rhabditida: Steinernematidae), and a fly, *Ormia depleta* (Diptera: Tachinidae). The nematode was applied as a biopesticide in many counties but spreads slowly. The wasp has spread to >15 counties after release in northern Florida. The fly spread fastest, now occupies southern and central Florida, but is limited by inability to overwinter north of »29^{o}N. Trapping stations in northern Florida established in 1979 showed no trend in total annual catches of mole crickets for nine pre-release years. After the biological control agents reached the vicinity of the trapping stations, numbers began to decline. They have declined for 12+ years. In 2003, the total annual catch of the worst of the three pests (*Scapteriscus vicinus*) achieved less than 0.5% of the annual average catch for the pre-release years. Statewide control is within reach.

The potential for a classical biological control programme against painted apple moth (*Teia anartoides* Walker) in New Zealand.

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The painted apple moth *Teia anartoides* Walker, a minor native Australian pest, is the target of an eradication programme in Auckland, New Zealand, where it poses a serious threat to indigenous and modified environmentsAs a precursor to a possible classical biocontrol programme in the event of eradication failure, searches for natural enemies of *T. anartoides* were carried out in autumn and in late spring to early summer 2003 in south-east Australia. At least 18 species of egg, larval and larval-pupal parasitoids were reared, the majority being Hymenoptera from the families Braconidae, Ichneumonidae and Eulophidae, and Diptera (at least two species of Tachinidae). The most common species from Victoria and Tasmania was *Cotesia* sp. (Hymenoptera: Braconidae), a solitary parasitoid of young larvae. The parasitoid guild showed greater diversity in spring-early summer than in autumn, and had some structural similarity with guilds of other Lymantriidae around the world. The guild was compared with existing natural enemies of *T. anartoides* in New Zealand and with those attacking other Lymantriidae occupying the same Australian habitats and host plants. Several Hymenoptera attacking *T. anartoides* larvae appear to be host specific and could act as effective biological control agents for this pest in New Zealand.

Prospects for biocontrol of mealybugs (Hemiptera:Pseudococcidae) in South African fruit orchards

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Mealybugs can cause extensive damage in South African fruit orchards and their presence on exported fruit may cause rejections if endemic species are present. We discuss the current status of natural biocontrol of the most important species, the effects of the commonly used insecticides on the commercially available parasitoid *Coccidoxenoides peregrinus* and the host range of this species. We conclude that there is good potential for natural biocontrol, especially if parasitoid numbers are augmented early in the season; that *Anagyrus pseudococci* is a good biocontrol candidate as generalist parasitoid; that most insecticides are disruptive to biocontrol agents, but that some insect growth regulators are biocontrol compatible; that *C. peregrinus* is selective on *Planococcus* spp. and therefore not useful for general mealybug control.

Pathology caused by infection of heliothine larvae with a small, midgut-specific, RNA virus.

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The tetravirus *Helicoverpa armigera* stunt virus (HaSV) is a small positive-stranded RNA virus infecting only midguts of heliothine caterpillars. Two genomic RNAs encode three proteins including a replicase and capsid protein. Transmitted horizontally *per os*, and also vertically, HaSV is acutely virulent towards very young larvae, causing rapid cessation of feeding, cessation of growth, and death. Older larvae show developmental resistance, even towards high doses. The complex pathology elicited by this simple virus has been studied using novel histopathology techniques and the genetic manipulation of HaSV to express reporter genes. Infection of young larvae leads to rapid virus spread throughout the midgut. Infected cells are rejected from the midgut epithelium, whose integrity is degraded despite a phase of rapid cell regeneration. In older larvae, infection foci are established but disappear as the virus is cleared from the midgut, in contrast to their spread in younger larvae. HaSV's efficacy and specificity give it significant potential as a biocontrol agent, as demonstrated in recent field trials. The capsid's simplicity led to the study of novel approaches for pest control involving virus production in non-host systems. Assembly of infectious HaSV was achieved in plant cells upon transfection of plasmids expressing viral RNAs and capsid protein.

Using transgenic maize in a prescriptive integrated pest management program

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Unlike the use of transgenic insecticidal cultivars for European corn borer (*Ostrinia nubilalis* Hübner), management, the use of transgenic hybrids for corn rootworms, *Diabrotica* spp., could work in concert with existing scouting programs and established economic thresholds. In essence, by monitoring their fields for corn rootworm adults in late summer, farmers could base their decision to use transgenic corn rootworm hybrids the following spring upon scouting input and knowledge of thresholds. Crop consultants and other essionals in the agribusiness sector could take a very active role in this decision-making process. Nearly half the continuous (non-rotated) cornfields in Illinois fail to support economic infestations of corn rootworm larvae. If only those fields judged to be infested with economic densities were planted to transgenic cultivars, in effect, a rather large refuge across the landscape could be created. Arguably, this would lessen the selection pressure for resistance development. In contrast to soil insecticides, transgenic insecticidal cultivars for corn rootworms will likely place greater selection pressure on the corn rootworm population. Important differences exist between a banded application of a granular soil insecticide and transgenic hybrids from a selection pressure perspective, and on this basis, a prescriptive approach for transgenic cultivars appears more justified.

The assassin bug, *Pristhsancus plagipennis* (Walker) (Hemiptera Reduviidae) – a potential biocontrol agent for IPM

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The Australian reduviid, *Pristhesancus plagipennis* (Walker), is a predator of Hemipteran and Lepidopteran pest insects in a range of cropping systems. As with many reduviid species, its biological control potential has been largely ignored because of low abundance in cropping areas coupled with the challenge of augmenting it for pest management purposes. This presentation outlines our research from the perspective of overcoming the barrier of augmenting *P. plagipennis* for biological control. A basic rearing methodology has been developed along with release techniques for field crops. Field studies on this predator have focussed on the biological control of *Helicoverpa* spp. (Lepidoptera: Noctuidae) and *Creontiades dilutus* (Hemiptera: Miridae) as well as the side effects of insecticides likely to be used within an IPM program. Both laboratory and field work indicate that *P. plagipennis* holds considerable promise as an applied biological control agent. However, to realise its full potential and be used commercially, the cost-effectiveness of the mass-production system needs to be optimised. Rearing technology options and their prospective impact on the feasibility of this predator will be discussed.

Advances in protein marking for mark-release-recapture and mark-capture studies

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We describe recent advances toward improving protein marking procedures for mark-release-recapture (MRR) and mark-capture studies. For MRR studies, a series of tests were conducted to determine if the sensitivity of an established rabbit immunoglobulin (IgG)-specific ELISA could be improved for detecting protein marks on insects. Five variations of ELISA were examined for their ability to detect rabbit IgG-marked *Hippodamia convergens*. The sandwich ELISA format was the most sensitive immunoassay. ELISAs with immunoreagent incubation intervals held constant at 5, 10, 20, or 60 minutes were examined for their ability to detect rabbit IgG-marked beetles. Results showed that the ELISA with 60 minute incubations yielded significantly higher ELISA readings. For mark-capture studies, inexpensive protein markers are being developed for studying landscape-level movement patterns of insects. Our approach is to utilize ELISAs that detect inexpensive proteins such as egg white, non-fat dry milk, and soy milk applied directly in the field using conventional insecticide spray rig equipment. The ELISA used to detect each protein is highly specific and sensitive, facilitating the use of multiple marks for studying various aspects of intercrop dispersal. The benefits and challenges of protein marking for MRR and mark-capture studies will be discussed.

Host-parasitoid dynamics in fragmented landscapes: patterns of dispersal and genetic variation

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The Glanville fritillary butterfly (*Melitaea cinxia*) has a large metapopulation in the Åland Islands in Finland and is attacked by two specialist parasitoids, *Cotesia melitaearum* (Braconidae) and *Hyposoter horticola* (Ichneumonidae). The two parasitoids have a striking difference in their range of dispersal, which is shorter than in the host butterfly in *C. melitaearum* but longer than in the host butterfly in *H. horticola.* Because of this difference in dispersal, and other differences in the biology of the parasitoids, *C. melitaearum* exhibits classic host-parasitoid metapopulation dynamics whereas *H. horticola* experiences the host metapopulation as a more continuous though patchy population. The genetic spatial structures of the two parasitoids, investigated with microsatellite markers, show a corresponding difference, with clusters of multilocus genotypes corresponding to the microgeographical population structure in *C. melitaearum* but not in *H. horticola.*

Development of microbial insecticides and their integration into farming systems in Australia.

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Biopesticides based on insect pathogens have enjoyed significant success in mainstream broadacre cropping in Australia. A number of commercial products based on nucleopolyheoviruses (NPVs) (Baculoviridae) have been registered for control of *Helicoverpa* sp. Primarily introduced for use in sorghum, NPVs have been widely adopted in cotton and other crops in IPM and insecticide resistance management. Consistent availability and quality of product, initially from the USA, has allowed the development of a significant market and subsequent development of new products produced in Australia supported by our research. NPVs are effective in controlling *H. armigera* that are resistant to chemical insecticides, with minimal disruption of natural enemies. Our work has shown significant improvements in performance of NPVs through formulation and application, and has established a baseline of susceptibility to NPVs in field populations across Queensland that suggests that there is little threat of resistance to these biopesticides. The success of biopesticides in Australian IPM has prompted our research and development of new potential products against *Helicoverpa*, mirids, pentatomids, diamondback moth and aphids. We will present our results on the identification, production, formulation, field-testing and integration into IPM of biopesticides based on entomopathogenic fungi and baculoviruses against these key pests.

A review of the surveys for natural enemies of *Mimosa pigra*: what does it tell us about surveys for broadly distributed hosts?

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The weed *Mimosa pigra* has been the target of a long running biocontrol project. This woody legume occurs naturally in all tropical American countries from Mexico to Argentina as well as many Caribbean islands. Surveys of natural enemies were made in seven countries and revealed a total of c. 420 insects and 5 fungi. This review focuses on assessing the collection effort in relation to the natural distribution of the host. We show species accumulation curves for some sites to estimate the number of visits required to find most species at a site. We examine the distribution of the agents where that information is available and compare it to the host distribution, to enable the prediction of the number of species that our surveys missed. We draw general conclusion that are applicable to biocontrol surveys of any host that occurs over a wide geographic range.

Inundative releases of *Trichogramma evanescens* **against the olive (***Prays oleae***) and jasmine (***Palpita unionalis***) moths**

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Field releases of mass-reared egg parasitoid were applied against the flower and fruit generations of the olive moth, *P. oleae* and the permanent presence of the jasmine moth, *P. unionalis* during two successive olive seasons of 2002 & 2003. The number of male moths captured in sex pheromone traps in treated plots was significantly lower than untreated one. Parasitism in eggs of target pests was higher in treated sites than control site. In the latter, parasitism on *P. unionalis* was mostly encountered than on *P. oleae* eggs. This is may possibly due to the activity of a *Trichogramma* strain other than *T. evanescens*. The strain is a theletokous species, a little bit darker than *T. cacoeciae*. It was bred from eggs of both moth species. The percentage of infestation of fruits that fell by *P. oleae* was generally higher in control site than treated one. Fruit harvest/tree was significantly greater in treated plots than control one. The outcome of inundative releases of *Trichogramma* wasps was partially successful and could provide a model for control of lepidopterous pests of olive trees, especially when the endemic species is used. When the right adapted candidate of wasp would be reached, it should be taken in consideration the right time and dose of each release, the emergence time of adult wasps and duration of activity of wasps under field conditions.

Feeding status of the parasitoid *Diadegma semiclausum* affects biological control of *Plutella xylostella*: implications for pest management.

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In Australia the solitary larval parasitoid, *Diadegma semiclausum*, a specialist on diamondback moth *Plutella xylostella* (DBM) has established as biological control agent. In field cages, fed and starved individuals of this parasitoid species foraging for hosts have been shown to have patch-leaving tendencies that differ considerably in host patches with the same host density. For this species, the movements in the field were continuously observed. The patterns emerging from these and other behavioural studies are built into a spatially explicit model to quantify the difference in the pattern of parasitism, when parasitoids are starved or fully fed. In the model, we mimic the spatial structure in Australian fields where year-round broccoli (*Brassica oleracea* var. *italica*) is planted. A cohort of new plants starts every fortnight. For studying how the physiological differences affect the parasitism levels in the field and therewith the biological pest control of DBM we used an individual based, spatially explicit, simulation model. We discuss the implications for biological control of DBM by relating the difference in longevity and the variation in foraging behaviour of the parasitoids due to different physiological state (fed versus starved) to variation in parasitism.

Competition and cannibalism in the gregarious egg parasitoid *Trichogramma carverae*: time-lapse imaging of development.

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Wasps of the genus *Trichogramma* have gregarious larvae that develop within lepidopteran host eggs. We address the question of whether gregarious larvae are completely tolerant of each other, and competition is solely due to a scramble for food, or whether physical attack occurs. Video microscopy was used to record feeding behaviour of larval *Trichogramma carverae* at different densities in *Epiphyas postvittana* eggs. Single larvae show a rapid food ingestion phase, followed by a period of extensive saliva release. The larva completely consumes the egg contents then extracts excess moisture from the egg before pupating. When multiple larvae are present, they rapidly consume all of the egg contents. Subsequently, further growth occurs by larvae feeding upon each other. The process continues until the surviving larvae reach a developmental stage beyond which competitive interactions seem to be no longer effective. Thus the number of larvae remaining at the end of the rapid ingestion stage dictates how many will survive to emerge as adults. We conclude that larvae are tolerant of siblings at low density if there is sufficient food for all to reach the threshold developmental stage. If not, their physical proximity results in attack and consumption of siblings.

Choosy egg parasitoids: specificity of response of egg parasitoids to plant volatiles induced by egg deposition

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Plants may respond to insect egg deposition by release of volatiles which attract egg parasitoids (1). Recently Scots pine (*Pinus sylvestris*) was shown to respond to egg deposition by the pine sawfly *Diprion pini*. Pine twigs with host eggs emit volatiles that attract the eulophid egg parasitoid *Chrysonotomyia ruforum,* a species specialized on diprionid eggs (2, 3). We tested whether this response is specific for the plant and herbivore species. *C. ruforum* shows a high specificity for oviposition-induced plant volatiles with respect both to the plant and herbivore species. Furthermore, the parasitoid´s response to these volatiles depends on its experience prior to the bioassay. While numerous generalist parasitoids are well-known to learn a wide range of volatiles when they had the opportunity to associate them positively with a host , the specialist *C. ruforum* was shown just to be able to learn specific volatile blends.

Patch use by egg parasitoids: theory, facts, and biological control

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This is a critical review of theoretical and empirical studies on patch use by egg parasitoids, determining if theory explains observed facts. It explores the possibility that egg parasitoids aggregate in host patches of high density or large size because classical patch-use theory predicts that foraging parasitoids should spend a relatively longer amount of time in high-quality patches, and the possibility that parasitoid aggregation results in density dependent parasitism. It stresses the fact that host age oundly influences patch use by parasitoids of eggs laid in masses because host quality declines with host age; this occurs simultaneously in all hosts in a patch (i.e. egg mass). When parasitoid females discover low-quality host patches just before host hatching, they should leave the patches as soon as possible to search for another patch. The case of *Anagrus sophiae* (= *delicatus*) females that attack only a small fraction of hosts in an egg mass and leave high-quality patches still carrying apparently mature eggs (Cronin & Strong, 1993) seems to be exceptional among quasi-gregarious egg parasitoids. Characteristics of patch use by egg parasitoids are discussed in relation to their use in biological control.

Selecting arthropod biological control agents: improving agent selection to minimize non-target impacts and maximize target impacts

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Non-target impacts and establishment of ineffective arthropod biological control agents targeting arthropod pests are receiving increased levels of research either through retrospective studies or via *a priori* evaluations before release to from quarantine. Glassy-winged sharpshooter, *Homalodisca coagulata*, an invasive xylophagous hemipteran in California, is the focus of a major classical biological control program with mymarid parasitoids that have been recruited from the pest's home range in the southeast U.S.A. Native sharpshooters potentially at risk from exotic parasitoids are being evaluated for susceptibility to attack as are native sharpshooters in French Polynesia, where *H. coagulata* established *circa* 1999. Laboratory studies assessing risk and parasitoid efficacy have indicated that screening parasitoids for potential efficacy is strongly influenced by parasitoid species and experimental conditions such as the complexity of the environment searched and relative host abundance provide. These different metrics for evaluating parasitoid efficacy can provide different answers as to which parasitoids will be most effective. Our work suggests that greater thought needs to be given to prevailing pest densities at the start of the biological project and how host-parasitoid dynamics will change as successful agents control the target pest making it less common. Lower pest densities because of successful biological control may affect which species or guilds of parasitoids come to dominate and regulate the system at low host densities compared to observations at the start of the program where pest numbers were higher.

Attractants for *Aleochara* spp. (Coleoptera: Staphylinidae), natural enemies of *Delia radicum* (Diptera: Anthomyiidae), in canola

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Defatted mustard seed meal has been shown to attract *Aleochara* species in vegetable brassicas in Sweden, and crop damage was reduced in plots with this treatment (Ahlström-Olsson & Jonasson, I0BC/WPRS Bull. 1992; 15:171). In trials with mustard flour in Canada and with defatted mustard meal in Switzerland, pitfall traps in canola plots with mustard preparations spread at 20 g/m² did not catch higher numbers of *Aleochara bilineata* than did traps in untreated control plots. However, in the Swiss trial, *A. bipustulata* catches in traps in mustard meal plots were almost 2.5 times those in control plots. Parasitism of *Delia radicum* pupae by *A. bipustulata* was significantly higher in mustard meal plots than in control plots, but parasitism by *A. bilineata* was not affected by the treatment. In Y-tube olfactometer studies, *A. bipustulata* exhibited preference for mustard meal compared to a blank control, but *A. bilineata* showed no preference. We are conducting further olfactometer studies comparing mustard meal and mustard flour in the presence and absence of soil and of moisture. Additionally, we are comparing the chemical composition of volatiles from potential attractants using gas chromatograph mass spectrometry.

Influence of host density on the dispersal of a specialist parasitoid

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The use of genetically modified corn in the United States to control the European corn borer, *Ostrinia nubilalis*, is creating a amatic shift in the spatial dispersion of hosts available to the specialist parasitoid *Macrocentrus grandii*. The persistence of this parasitoid in the Midwestern corn belt may be limited by its dispersal abilities, and its behavioral response to large regions of low host densities created by the widespread use of genetically modified corn. We marked parasitoids developing on *O. nubilalis* using foliar applications of rubidium chloride to 0.2 ha corn plots. Marked parasitoids were presented with transgenic (low host density) and nontransgenic (high host density) corn fields and their dispersal in the two types of fields was monitored using sticky traps. More than 10,000 parasitoids were captured in the first year and preliminary results show differences in the behavior of male and female *M. grandii* dispersing into transgenic corn fields. Parasitoids were captured throughout the 8 ha transgenic corn fields demonstrating that dispersal into low host density fields is possible at this scale. Rubidium levels in parasitoids collected from the 0.2 ha treated plots were 8 times higher than in unmarked parasitoids.

Biological control of *Bemisia tabaci* **by predators**

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The sweetpotato whitefly, *Bemisia tabasi* (Gennadius), is a widespread pest of crops in the world, which recently seriously occurrs in China, and damages the most economically significant crops of cotton, vegetables and ornamentals, showing a tendency of annual outbreaks with the trouble problem urgent to be solved in agricultural productions. The predatory enemies of *Bemisia tabasi* in China have some species of ladybeetles, thrips, aphids lions and predatory mites, of which the ladybeetles *Serangium japonicum* and *Delphastus catalinae* are active as the dominant species. Both the two species of ladybeetles have a good efficiency feeding on the different stages of *Bemisia tabasi*, especially preferring to the whitefly eggs. At the laboratory, *Delphastus catalinae* develops a generation by 17-22 days at 25-28°C, with the ladybeetble population increases 21.61 times more than that of the fore generation, and develops about 15 generations per year. The predatory quantity per day reaches up 1198 and 670 of whitefly eggs fed by a 4th instar larva and an adult female of *Delphastus catalinae* respectively. The mass rearing of ladybeetbles carries out followed by the rearing process of host plant – host whitefly – ladybeetble.

The Sirex woodwasp control programme in South Africa

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The Sirex woodwasp, *Sirex noctilio*, was first detected in South Africa in the Tokai plantation, Cape Town in 1994. *Sirex noctilio*, together with its symbiotic fungus *Amylostereum areolatum*, inhabits and kills pine trees. Since its initial detection in South Africa, this exotic pest has spread rapidly and is causing increasing losses to the pine industry. Three biological control agents were introduced into South Africa soon after *S. noctilio* was first detected. These included the parasitic nematode, *Beddingia siricidicola*, which is considered to be the best biological control agent against the woodwasp. Unfortunately, the biocontrol agents have not spread together with *S. noctilio*. To address this problem, *B. siricidicola* has been re-introduced on a much larger scale and on a more continuous basis than was previously attempted. The aim here has been to ensure that the nematode becomes established and that it achieves sufficient control of the wasp in South African pine plantations. Studies are also underway to investigate possible differences in the life-cycle of *S. noctilio* in different climatic zones of South Africa. Furthermore, we are considering evidence to suggest that the wasp has been introduced into the country more than once.

Associative learning of plant chemicals by the egg-larval parasitoid, *Ascogaster reticulata* **Watanabe (Hymenoptera: Braconidae)**

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We examined the learning response to authentic chemicals in *Ascogaster reticulata* Watanabe, an egg-larval parasitoid of the smaller tea tortrix, *Adoxophyes honmai* Yasuda (Lepidoptera: Tortricidae). After 3 times' conditionings of chemicals associated with the host egg mass, the strength of the learned response in a female was evaluated by the distance walked along the chemical cues with her characteristic antennating response in 9-cm Petri dish assays. We compared the learned response of females to 5 authentic chemicals, i.e., linalool, geraniol, methylsalicylate, *(Z)*-3-hexenol and benzyl alcohol. The order of learned responses was as follows: linalool = geraniol > methylsalicylate > *(Z)*-3-hexenol = benzyl alcohol. When the females were conditioned with geraniol, they responded to geraniol significantly longer in a choice between geraniol and linalool. The linalool-conditioned females positively chose linalool in the same choice situation, but not significant. Use of the mixtures of these chemicals in different ratio also had similar effect.

Augmentation of parasitoids by mass-hatching devices: a new biocontrol technique

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Augmentation can be an effective biocontrol strategy to increase the impact of antagonists. However, mass-reared beneficials are generally expensive and often poorly adapted to field conditions. The goal of this study was the development of a device, which selectively permits parasitoids to emerge out of pest-infested plant material. Plant material containing the host and its parasitoids can be collected and stored in large closed devices from which the generally smaller parasitoids can emerge through a selective tissue filter of appropriate mesh size. We used mass-hatching devices to augment populations of indigenous parasitoids of the invasive horse chestnut leafminer, *Cameraria ohridella* (Lepidoptera, Gracillariidae). Whereas more than 75 % of the 15'000 parasitoids passed the separator only 1 % of the 50'000 leafminers overcame the tissue filter. Furthermore, the set-up of mass-hatching devices in the city of Basel, Switzerland, significantly increased parasitism rates of *C. ohridella*. Mass-hatching devices may thus contribute to an integrated control strategy against *C. ohridella* in Europe. In general, mass-hatching devices are a promising new augmentative approach to control insect pests with a dormant stage in or on plant residues and should be considered as a supplementary, cost-effective biocontrol technique.

Molecular genetic investigation of local adaptation and incipient speciation in a tritrophic system

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Seven polymorphic microsatellite loci were assayed in two groups of the insect parasitoid, *Cotesia congregata* (Say) [Hymenoptera: Braconidae]. One group originated from *Manduca sexta* L. and *Manduca quinquemaculata* (Haworth) on tomato and tobacco ("solanaceous group") and the other from *Ceratomia catalpae* (Boisduval) on catalpa ("catalpa group"). Allelic distributions differed significantly between the solanaceous and catalpa groups (Fisher's exact test $P = 0$ overall and for each locus). Across all loci, F_{ST} was < 0 for within-group comparisons and > 0.47 for between-group comparisons. Wasps from the solanaceous and catalpa groups were examined for mtDNA sequence variation to investigate their possible status as sibling species. One conserved mtDNA gene (16S) showed a single fixed nucleotide difference between the two groups. The results support the hypothesis that host plant association is a significant factor influencing population structure of *C. congregata*.

Selecting arthropod biological control agents against arthropod pests: how can the science be improved to decrease the risk of releasing ineffective agents?

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From the practitioner's viewpoint, there are several principles that, if observed, will improve the chances for successful agent selection:

1) Involvement of morphological and/or molecular characterizations is needed from the inception of the project. When the native range/origin of a pest is unknown, this will help to identify significant differences in pest populations and may reveal their actual native range. Identification and characterization of the natural enemy geographic populations and possible host range differences is equally important.
2) Use of climate matching tools is a good starting point for locating climatically-adapted populations of natural enemies.
3) Native range studies should be conducted to evaluate the impact of exotic agents on the target pests.
4) Concurrent and parallel studies should be conducted in the potential release area to document gaps in native enemy activity or other ecological factors that contribute to high pest populations. This also means that a committed practitioner is in place in the receiving area.
5) Collection of natural enemies of species that are taxonomic relatives or ecological homologues of the target pest and which occur in the same or neighboring habitats may reveal field specificities.
6) Quarantine evaluations can provide a preliminary screening of potential effectiveness.
7) Field cage/semi field evaluations are desirable to validate the results of preliminary screenings under more realistic conditions.

These approaches will be discussed in relation to examples of recent projects.

Management of sucking pests in broadacre crops using *Beauveria bassiana* and *Metarhizium anisopliae*.

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Biopesticides based on insect pathogens have been widely applied in an integrated pest management (IPM) strategy against *Helicoverpa* species in Australia, maintaining beneficial insect populations and minimising the development of resistance to chemical insecticides. As reliance on chemical control has reduced in cotton and grain crops, sucking pests such as mirids (*Creontiades* sp.) and green vegetable bug (*Nezara viridula*) are emerging as significant problems. Application of broad-spectrum insecticides against sucking pests threatens the successful IPM strategy by destabilising natural enemy populations and triggering outbreaks of *Helicoverpa sp,* however there are currently no selective insecticides registered for sucking pests. We have identified *Beauveria bassiana* and *Metarhizium anisopliae* isolates active against these pests. Field trials demonstrated control of mirids equal to that achieved with the chemical Dimethoate by four to six days after treatment for both isolates at 1 x 10^{13} spores/hectare. There was no significant impact on natural enemies in any of the field trials. Monitoring has indicated that sucking pest populations build up in pulse crops through the vegetative stage. We are modelling pest populations and developing a pest management strategy based on population management using biopesticides as an alternative to reactive knockdown using broad-spectrum chemical insecticides.

Red imported fire ant: benefit or detriment to conservation biological control in cotton?

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Predation of sentinel eggs of *Helicoverpa zea* and *Spodoptera exigua* (Noctuidae) was 1.5-2.0 and 4.1 times greater, respectively, in the presence of red imported fire ant, *Solenopsis invicta* (Formicidae), than when fire ants were excluded from 3-ha plots of cotton in central Texas in 2001-2002. Most (68%) of the observed predation events on beet armyworm eggs was due to *S. invicta*. Densities of cotton aphid, *Aphis gossypii*, were 5.5 times greater in the presence of fire ants compared to ant exclusion plots, although aphid densities did not reach economic levels. In additional field trials conducted in 2003, densities of cotton aphid were not significantly different between ant treatments and again did not exceed the economic threshold (50 aphids/leaf). Densities of *Hippodamia convergens* (Coccinellidae) adults and larvae were significantly greater in the ant exclusion treatment Red imported fire ants are intraguild predators in cotton. However, our results suggest that the overall impact of the red imported fire ant is beneficial to cotton pest management in Texas and efforts to enhance fire ant densities have the potential of increasing biological control of nocutid pests.

Oral toxicity of *Photorhabdus* bacterial toxins against thrips species

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Photorhabdus and Xenorhabdus bacteria are symbiotic to entomopathogenic nematodes. These bacteria are released by nematodes after infection of insects. The oral toxicity of excretion products of several Photorhabdus and Xenorhabdus strains was tested on two thrips species: Frankliniella occidentalis and Thrips tabaci. Out of 46 Photorhabdus isolates and 6 Xenorhabdus isolates only 6 North American P. temperata isolates were toxic to the thrips species. After 7 days of inking from P. temperata supernatant a mortality of 90% was recorded and reproduction was inhibited. Thrips were also killed after feeding from leaves that were treated with the toxins. Possibilities of using P. temperata for the control of thrips will be discussed.

Understanding target weed population dynamics can improve our ability to select effective agents against weeds

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Understanding the population dynamics of a weed and the role of environmental factors in influencing these dynamics can help us prioritise insect and pathogen guilds in terms of their likely efficacy in controlling target weeds. In order to be effective, an agent needs to inflict sufficient damage, on appropriate organs or lifestages, and do so in a timely manner. Modelling the population dynamics of a weed can indicate the likelihood of agents from different guilds being able to inflict these desirable damage patterns under the range of environmental conditions that are likely to be encountered in the introduced range. The most desirable agents are those that, when released from their own predators or parasitoids, would act upon the plant population in the manner that most amatically reduces the problems caused by the weed – characteristics that will vary from weed to weed. The characteristics of the most desirable agents can then be used to influence the suite of agents that are specifically targeted for searching, and to help prioritise them for importation and host-specificity testing. If agent establishment follows the lottery model, then prioritising agents that, should they establish, are most likely to be effective should improve our overall success rate.

Efficacy of nuclear polyheosis virus with azadirachtin on the mortality, feeding, biological, biochemical and histopathological profiles of *Helicoverpa armigera.*

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The mortality of third instar larvae of *Helicoverpa armigera* larvae was observed in a dose dependent manner and the time to kill was significantly reduced in the combined treatment of Nuclear Polyheosis Virus (NPV) and Azadirachtin (AZA). The average leaf disc consumption of *H. armigera* larva fed with NPV (5 x 10^4 PIB/ml) treatment was 1.90 cm^2/larva and AZA treatment (4 μg/cm^2) was 0.63 cm^2/larva whereas it was significantly reduced to 0.21 cm^2/larva in the low dose combined treatment. Significant reduction in the larval and pupal weights, elongation of larval and pupal duration and a astic reduction in the adult longevity, fecundity and survivability were observed after the combined treatment. Reduced amounts of protein (35.8 mg/g), lipid (47.4 mg/g), carbohydrate (35.7 mg/g), amino acids (32.6 mg/g), DNA at 96 hrs post-treatment (82 µg/g) and RNA at 96 hrs post-treatment (2.4 µg/g) were observed after NPV and AZA treatment. These biochemical changes suggest that AZA did not affect the multiplication of NPV and further there is a synergistic interaction between NPV and azadirachtin. The combined treatment caused a severe necrosis of the midgut epithelial cells and loss of cell shape. The combined treatment of NPV and azadirachtin had much greater effect in relation to the pest management strategy.

Suitability of parasitized greenbugs (*Schizaphis graminum*) as food source for predatory Coccinellidae.

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Laboratory studies were conducted to determine first, the stage of development at which the predatory coccinellidae, *Hippodamia convergens*, *Coccinella septempunctata* and *Coleomegilla maculata* are able to start feeding on the parasitized greenbugs "mummies" (*Schizaphis graminum*), and second to determine the nutritional effects of mummies on the growth, development and survival of *Coccinella septempunctata*. Lady beetles were collected from the field and fed unlimited amount of pea aphids (*Acyrthosiphon pisum*) until when they laid eggs. A batch of coccinellid larvae at each instar was fed fresh mummies on daily basis and observed. The preliminary results showed that *Hippodamia convergens* start feeding on mummies at the third instar, even though *Coccinella septempunctata* start feeding on mummies at the second instar, and each instar larva was able to molt to the next stage of development. Further results of the experiment show that *C.septempunctata* take long to develop and they survive through one molt when they are fed mummies. More results will be obtained at a later stage of the study. This study confirms that *Coccinella septempunctata* and *Hippodamia convergens* are able to feed on the parasitized greenbugs at both larval and adult stages of development.

Isolation and characterization of novel insecticidal *cry1*-type genes from *Bacillus thuringiensis* strains

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To identify new *cry1*-type insecticidal genes, 56 lepidopteran-specific *Bacillus thuringiensis* (Bt) strains were isolated from Korea. Universal oligonucleotide primers, K5un2 and K3un2, were designed and used to amplify all known *cry1*-type genes with PCR from 56 Bt isolates. The restriction fragment length polymorphism (RFLP) patterns of the PCR-amplified fragments revealed 7 distinct patterns, and a standard isolate was selected from each one of the RFLP groups. Through cloning and sequence analysis of the PCR-amplified fragments showing unique RFLP pattern, 7 novel *cry1*-type genes were identified. Bt K-1 and Bt 2385-1 strains were selected to isolate and characterize the active regions of the novel genes, and then 5 novel genes were cloned from them. To verify the activity against lepidopteran larvae, the novel genes were expressed using baculovirus expression vector system and the bioassay was performed to *Plutella xylostella* (Px) and *Spodoptera exigua* (Se). All novel *cry1*-type genes had high toxicities against Px larvae and, *cry1-44* and *cry1-5* had the highest toxicity against Px and Se larvae, respectively. Also, *cry1-1* and *cry1-5* had dual toxicities against Px and Se larvae. Finally, each novel gene must be a very useful resource for development of microbial insecticide and insect resistant plants.

Bats feed on crop pests in California's Sacramento Valley

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Food habits of Mexican free-tailed and Yuma bats were examined in the Sacramento Valley in California. Analyses of guano samples indicated that these two species of bats fed on moths, beetles, and plant bugs suggesting they might help control crop pests. Further studies in cotton, walnuts, pears, and alfalfa showed that bats forage extensively in these crops; however, their impact on biological pest control remains unclear. A colony of 150 bats will feed on more than a million insects a year. In addition, bats may protect crops by their echolocation calls alone. Many insects, including moths, are sensitive to bat calls and take evasive action when they detect bats by turning or diving to the ground, which may disrupt their mating and egg-laying behavior. Given the beneficial nature of bats, growers in California have been putting up bat houses to attract bats to their farms for pest control. Analysis of 175 bat houses in California showed that bats prefer houses that were attached to wooden barns, at least ten feet off the ground, that received morning sun and afternoon shade.

What hypervariable molecular markers tell us about parasitoid population structure and dynamics at both landscape and geographic scales

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We here discuss the use of polymorphic microsatellite markers to study natural populations of the hymenopterous primary parasitoid, *Diaeretiella rapae* (M'Intosh), a polyphagous parasite of crucifer and cereal aphids. Using five such markers, its population structure and dynamics was examined at the field and landscape scale when attacking cabbage aphid, *Brevicoryne brassicae* (L.) infesting oilseed rape, *Brassica napus* at two sites in southern Britain. Parasitoid samples were collected at different spatial scales ranging from individual racemes to field corners. The data were analysed using Bayesian and other statistics to provide new insights into parasitoid movement, persistence and breeding structure. In addition, the same markers were later used to study the genetic variability of *D. rapae* at four geographic sites (Western Australia, France, Morocco and Kazakhstan) reared on the Russian wheat aphid (RWA), *Diuraphis noxia* (Mordvilko). When compared with Old World populations, Australian parasitoids had low allele length and frequency variation, suggesting a significant founder effect. Marked genetic differentiation between populations was also apparent, potentially important for host utilization in this species when introduced to a new geographic area/s. Low genetic variation and gene flow in a founder population could limit evolutionary potential, including the ability to switch to novel introduced hosts, e.g. RWA.

The density and diversity of parasitic Hymenoptera as bio-indicators of habitat disturbance in a cotton producing region

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The presence and diversity of hymenopteran parasitoids were compared in cotton with sprays of chemical insecticide, and protected with a bioinsecticide in Huehuetán, Chiapas, México from September, 1999 to January, 2002. Parasitoids were collected weekly from phytophagous insects on upper, middle and lower leaves, squares, flowers and bolls, held in styrofoam cups, with a semi-transparent lid. During the last two years, parasitoids were also collected by vacuuming above the plant canopy. Chemical insecticide decreased both the incidence and diversity of parasitoids early in the first year; however, the guild of natural enemies reappeared and became more abundant and diverse in the chemical treatment. This was associated with the protection brought to the crop by chemical insecticide sprays which resulted in more productive plants, furnishing more refuge to parasitoids late in the season. However, when the crop was rainfed, and phytophagous insects did not cause significant plant damage, natural enemies were affected by the chemical insecticide. Over 300 morphospecies of hymenopteran parasitoids were collected during the experiment. The fact that parasitic Hymenoptera occur at the top of the arthropod food chain makes this group of insects particularly useful in evaluating factors which control and often simplify natural and man-made ecosystems.

The impact of predatory bugs on *Helicoverpa armigera* in Australian cotton crops

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The bigeyed bug, *Geocoris lubra*, and the damsel bug, *Nabis kinbergii*, are two common insect predators in Australian cotton crops. These predators are known to feed on the lepidopteran pest, *Helicoverpa armigera*, however such predation is difficult to observe and quantify under natural conditions. The detection of specific protein markers in predator gut contents allows direct measurement of predation from field-collected individuals. Here, I outline the development of ELISA protocols to detect predation by these two species on *H. armigera* using a species-specific monoclonal antibody. Although prey-specific proteins were detectable for only short periods within the predators, positive examples of predation have been recorded. Adult and juvenile predators of both species were collected from commercial cotton farms in northern NSW during the 2001-02 and 2002-03 seasons. Approximately 12% of *G. lubra* and 24% of *N. kinbergii* tested positive for recent predation on *H. armigera*. Predation rates were similar in both seasons although the relative abundance of *H. armigera* was lower in 2001-02 than 2002-03. This suggests these insect predators can contribute to the suppression of *H. armigera* even when pest populations are low.

Viral pathogens for the control of painted apple moth *Teia anartoides* (Lepidoptera: Lymantriidae)

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The painted apple moth (PAM), *Teia anartoides* (Lepidoptera: Lymantriidae) is a native of Australia, where it is a horticultural pest. Recently, PAM was found in Auckland, New Zealand and poses a threat to horticulture, pine forests and native plants. An eradication campaign was mounted in the Auckland suburbs including aerial applications of the biopesticide Foray (based on *Bacillus thuringiensis*) and releasing sterile male moths. To supplement eradication, we have investigated the availability and potential of microbial insect pathogens to control PAM. Efforts to identify potential biological control agents have included evaluation of several nucleopolyheovirus (NPV) strains from related species (e.g. other lymantriids) and from commonly found forest or horticultural pest insects. Most were non-pathogenic or showed minor sub-lethal effects and one appeared to potentiate a virus present at low levels in PAM larvae. However, an NPV (*Oran*NPV), discovered in PAM in Australia in the 1960s, was highly virulent as were two NPV stains isolated from other lymantriid species (*Orygia pseudotsugata* and *O. leucostigma*). Finally, a cypovirus, isolated from PAM in New Zealand, was infective in bioassays. Preliminary sequence data suggests that this cypovirus is an undescribed member of the *Cypoviridae*. Host range testing of these viruses is underway.

Occurrence and abundance of spiders in cotton ecosystem of Tamil Nadu, India

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Agricultural entomologists have acknowledged the importance of spider species as a major factor in regulating pest populations of crops. Despite their importance were highlighted in various ecosystems by several workers, the role they play in regulation of pest populations in cotton ecosystem has received scant attention in India and few workers have attempted on faunistic survey in recording the spider fauna in cotton ecosystem. A survey was conducted at major cotton growing areas of Tamil Nadu *viz.,* Attur, Bhavanisagar and Coimbatore during winter cotton 1998 (July- December) and at Kovilpatty and Srivilliputur during summer cotton 1999 (February – June). The survey yielded 21 species *viz.*, *Araneus* sp., *Argiope catenulata* Doleschall*, A. minuta* Karsh, *Neoscona theisi* Walcknear, *Cyrptophora cicatrosa* Stoliczka, *C. viridanus* Cambridge, *Oxyopes javanus* Thorell, *O. rufisternum* Thorell, *Peucetia viridana* Stoliczka, *Plexippus paykulli* Audouin, *Tetragnatha javana* Thorell, *Thomisus cherapunjens* Tikader, *T. pugilis* Stoliczka, *Leucage decorata* Blackwall, *Gasterocantha citricola* Forskal, *Neoscona elliptica* Tikader, *Clubiona* nr. *assodes* Cambridge, *Lycosa pseudoannulata* Boes. et Str., *Pardosa* sp., *Bianor* sp. and *Salticus* sp. grouped under 16 genera belonging to eight families (Araneidae, Argiopidae, Clubionidae, Oxyopidae, Lycosidae, Salticiade, Tetragnathidae and Thomisidae).

Genetic differentiation of hymenopteran parasitoids

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The ecology of parasitoids is strongly influenced by the host plant. Parasitoid fitness is affected by a variety of plant traits. Thus, it is reasonable to assume that adaptation to plant traits could promote phenotypic and genotypic differences among populations of parasitoids. We present data on phenotypic and genotypic differences of two braconid parasitoids ovipositing on the same host on different host-plants. We compare the developmental time, percent parasitism, and adult weight of a generalist (*Cotesia marginiventris* (Cresson)) and of a specialist parasitoid (*Aleiodes nolophanae* (Ashmead)) of the green cloverworm and use AFLP data as a measure of genotypic differences in parasitoids emerging from hosts on alfalfa versus those reared from hosts on soybeans. AFLPs have been widely used in ecology to explore genetic differentiation among populations in several species and cluster analysis methods have been applied to visualize these differences and to test all sort of hypotheses. However, these analyses are potentially flawed because there is no agreed upon criteria for determining the appropriate number of individuals and bands for such representations to capture variation in the field. We present a method for determining the number of individuals to collect and number of bands to consider when studying genetic differentiation.

Habitat manipulation for insect pest management in Australian cotton cropping systems

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Adoption of within-field monocultures of cotton crops in Australia discriminate against and reduce the establishment and activity of natural enemies of pests because they lack ecological diversity. Increased habitat diversity in the cotton cropping systems has been achieved by planting refuge crops adjacent to cotton fields or as strips or centrally located blocks. The use of refuge crops in this way has enhanced (1) the conservation and establishment of natural enemies on cotton crops early in the cotton season to suppress pest populations and (2) the diversion of pests away from cotton onto the refuge crops resulting in the reduction of pest population. Interplanting cotton crops with refuge crops such as lucerne, the strips should be 8-12 metres wide and 300 metres apart and should run the whole length of the adjacent cotton field. The problem associated with the use of refuge crops for pest management in cotton is that natural enemies may not be inclined to move from the refuge crops to forage the adjacent cotton crops. The movement of the natural enemies can be improved by either applying a natural enemy food attractant to the cotton crops or by reducing the size of the strips planted to refuge crops. The criteria for the selection and management of the refuge crops as well as their deployment in IPM programs in cotton will be discussed.

Selecting parasitoids for biological control introductions: codling moth as a case study

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The parasitoid assemblages of lepidopteran pests are more diverse and interactive than those of homopteran pests, posing a greater dilemma for the biological control practitioner in selecting candidates for introduction. How do we identify the single best candidate for introduction, or if more than one species is selected, how important is it that they show no direct interactions? The recent biological control program against codling moth in California will be used to illustrate both theoretical and practical aspects of parasitoid selection for introduction. From a theoretical perspective, the importance of pest refuges from parasitism, antagonistic interactions, and parasitoid aggregation will be emphasized. In contrast, from a practical perspective, the importance of ease of collection, rearing success, and risk will be considered. These contrasting perspectives provide an interesting example of parasitoid selection as both an art and a science.

RESPONSE OF *TRICHOGRAMMA PLATNERI* AND *T. PRETIOSUM* TO PATCHES OF SINGLE AND CLUSTERED HOST EGGS

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COMMERCIALLY PRODUCED TRICHOGRAMMA SPECIES ARE WIDELY USED IN AUGMENTATIVE BIOLOGICAL CONTROL PROGRAMS. HERE WE TEST WHETHER TWO NORTH AMERICAN SPECIES *T. PLATNERI* AND *T. PRETIOSUM*, DIFFER IN THEIR RESPONSE TO AND EFFICIENCY OF USE OF SINGLE AND CLUSTERED HOST EGGS. THE OVIPOSITION BEHAVIOR OF INDIVIDUAL EXPERIENCED PARASITOID FEMALES WAS MONITORED CONTINUOUSLY IN PATCHES (3.5 X 3.5 CM CARD) WITH 16 SINGLE SPACED OR CLUSTERED EGGS OF *EPHESTIA KUEHNIELLA* AT 25°C FOR UP TO 1 H OR UNTIL THE FEMALE LEFT THE PATCH. THE OVIPOSITION BEHAVIOR OF EACH PARASITOID SPECIES IN EACH PATCH TYPE WAS SUMMARIZED AS GENERALIZED ETHOGRAMS, PATTERNS OF PATCH USE, TIME BUDGETS AND THE SEQUENCE OF SEX ALLOCATION. *T. PLATNERI* HAD A GREATER RATE OF OVIPOSITION IN PATCHES WITH SINGLE HOST EGGS, AND AN INDETERMINATE ALLOCATION OF SEXES IN PATCHES OF CLUSTERED HOST EGGS. IN CONTRAST, *T. PRETIOSUM* SHOWED BOTH A GREATER RATE OF OVIPOSITION AND A MORE EFFICIENT SEX ALLOCATION IN PATCHES OF CLUSTERED HOST EGGS. THE MORE EFFICIENT USE OF HOST EGG CLUSTERS BY *T. PRETIOSUM* THAN BY *T. PLATNERI* SUGGESTS THAT CLOSER ATTENTION SHOULD BE PAID TO MATCHING THE OVIPOSITION BEHAVIOR OF TRICHOGRAMMA SPECIES TO THE OVIPOSITION STRATEGY OF THE TARGET PEST.

Metarhizium anisopliae as a mycoinsecticide for taro beetles (*Papuana* spp.) in Papua New Guinea and other Pacific Islands.

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Taro is an important root crop for Melanesian and Polynesian people. It is extensively cultivated in PNG as well as Solomon Islands, Fiji, and Vanuatu. The adults of several species of Taro beetle are naturally occuring pests in PNG and the Solomons and recent introductions into other countries such as Fiji. The larvae are detritus feeders and are not an economic problem. The adults burrow down through the soil and eat large holes in the developing corm resulting in the death of the plant or a non-marketable product. An isolate (FI-1452) of *Metarhizium anisopliae* found on *P. woodlarkiana* in PNG is virulent for that species as well as for *P. heubneri* and *P. uninodis*. This isolate is being mass produced on rice in PNG and Fiji and tested in field trials for control of Taro beetle as part of an ACIAR/SPC project. Initial tests in PNG at 3 sites with doses ranging from 10g to 300g applied into the planting hole have increased the proportion of marketable corms from 33% to 65%. Research is still evaluating the most effective dose and an acceptable formulation. Also possible combinations with chemical pesticides are being studied.

Field population dynamics of egg parasitoids of the glassy-winged sharpshooter and smoke-tree sharpshooter in California

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The glassy-winged sharpshooter (GWSS), *Homalodisca coagulata* (Say) (Hemiptera: Cicadellidae), is under chemical management in California in the vicinity of Temecula, the Coachella Valley, the southern San Joaquin Valley, and Ventura Co. This insect is of concern not only because it vectors various strains of *Xylella fastidiosa* with potential to cause disease on a wide variety of crops (e.g., Pierce's disease on grapes) but also because control mandated to limit its spread in the state causes disruption of nursery shipments and movement of harvested citrus. The smoke-tree sharpshooter, *H. liturata* Ball, is native to California and has also been shown to vector *Xylella* diseases although apparently to a letter degree than the introduced *H. coagulata*. A two-year field study in the vicinities of Riverside and Desert Center, CA evaluated levels of egg parasitism on these two sharpshooter species across five fields containing a total of ten host plants. One of these fields was used by the California Deptartment of Food & Agriculture as a site for augmentative release of four exotic species/strains of parasitoids over the duration of this study. The temporal pattern of egg parasitism by eight species, six *Gonatocerus* (Hymenoptera: Mymaridae) and two *Ufens* (Hymenoptera: Trichogrammatidae) are discussed.

Potential biological control of invasive alien flatworms

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The British Isles have been invaded by antipodean terrestrial flatworms, termed locally the 'New Zealand flatworm' (*Arthurdendyus triangulatus*) and the 'Australian flatworm' (*Australoplana sanguinea*). Both species are earthworm predators and there is increasing evidence that *A. triangulatus* negatively affects earthworm populations, in particular anecic species of earthworm. Decline/alteration of earthworm populations could have damaging knock-on effects on soil fertility and other earthworm-feeding wildlife. However, control options are limited due to the flatworms' abundance and cryptic nature. An exclusion experiment was designed to investigate the impact of Carabidae on *A. triangulatus*. Barriered field plots, half of which were treated with insecticide, were sampled weekly for flatworms and carabids. In insecticide-treated plots, there were fewer carabids but more flatworms, compared to untreated controls. However, the difference between treatments was not large, suggesting that carabids moderate rather than control *A. triangulatus* populations. 'Classical' biological control is another option. A Tasmanian midge, *Planarivora insignis* (Mycetophilidae) is the only known parasitoid of terrestrial flatworms. Little is known about this midge beyond a single publication in 1965. Could this midge be used as a control agent for invasive flatworms?

Bollgard®II – Will it affect abundance of *Microplitis demolitor* in Australian cotton agroecosystems?

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The braconid *Microplitis demolitor* is the most important parasitoid of early instar *Helicoverpa* spp. in the cotton agroecosystem. Late first and second instar larvae are the preferred host stage for oviposition, with host death usually in the third or fourth instar. Feeding by parasitised larvae is greatly reduced, with diet consumption about 10% of that consumed by healthy larvae. In unsprayed non-Bt cotton, parasitism rates of second and third instar larvae often exceed 50% in the latter half of the growing season. *Microplitis demolitor* also transmits ascovirus when ovipositing which usually results in the death of the immature parasitoid in the ascovirus-affected host. Although routine insecticide use in conventional cotton can be disruptive to adults and immature stages of *M. demolitor*, the pupal case offers protection from most insecticides. Cotton is just one of many crop hosts that harbour *M. demolitor* in the farming system. Bt transgenic cotton, particularly Bollgard®II, results in extremely high mortality of larvae of *Helicoverpa* spp. It is not known to what extent the increased adoption of Bollgard®II will affect abundance of *M. demolitor* through reduced host availability in cotton.

Quantifying conservation biological control of *Bemisia tabaci* in the cotton system

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Multiple mortality forces act on insect populations. Estimating mortality caused by natural enemies within the context of other interacting mortality agents is important to understanding the contribution of biological control to pest suppression. Field studies examined the role of natural enemy conservation in cotton within the context of alternative management strategies for *Bemisia tabaci* based on selective and non-selective insecticides. *In situ* life tables were used to quantify and interpret the effects of natural enemies relative to other contemporaneous mortality factors acting on pest populations. Single applications of selective insecticides initially functioned by replacing some mortality from predation and parasitism, and contributing an immediate, and essential, level of irreplaceable mortality. However, because these selective materials did not disrupt natural enemies, parasitoids, and especially predators, were able to supply irreplaceable mortality that contributed to pest suppression for the remainder of the season. This extension in time also served to increase the cumulative chance of pest suppression by weather-related factors. In contrasted systems based on broad-spectrum insecticides, repeated applications were required for pest control. This study demonstrates the role of multiple mortality factors in managing *B. tabaci* and provides a mechanistic understanding of the important contribution of conservation biological control.

"The biology and characterisation of an ascovirus (Ascoviridae) from Australia"

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Ascoviruses (Ascoviridae) are a group of enveloped DNA viruses that cause a chronic and lethal disease in insects. Most of the described ascoviruses are hosted in the larval stage of the Noctuidae (Lepidoptera). Unpublished reports indicated that an ascovirus was found at high incidence (>50%) in populations of *Helicoverpa armigera* (Hübner) and *Helicoverpa punctigera* (Wallengren) in southeast Queensland, Australia. These pathogens were thought to be vectored by the braconid parasitoid wasp *Microplitis demolitor* (Wilkinson). I have formally identified and characterised this ascovirus by examining; the genetic relationship to other known ascoviruses (using RFLPs, Southern blot hybridisations and sequencing the polymerase gene), the host range of the virus and the histopathology. The Australian ascovirus was similar to the *Heliothis virescens* (Fabricius) ascovirus (HvAV) and the *Trichoplusia ni* (Hübner) ascovirus (TnAV) from the USA. To further understand the biology of the Australian ascovirus, I studied its transmission and interaction with hymenopteran parasitoids; I examined other "alternative" modes of transmission and its field prevalence in Australia. The Australian ascovirus appears to be an opportunistic pathogen; it may rely on multiple hosts, multiple vectors and multiple modes of transmission for dissemination and persistence in the field.

Choosy egg parasitoids: specificity of response of egg parasitoids to plant volatiles induced by egg deposition

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Plants may respond to insect egg deposition by release of volatiles which attract egg parasitoids (1). Recently Scots pine (*Pinus sylvestris*) was shown to respond to egg deposition by the pine sawfly *Diprion pini*. Pine twigs with host eggs emit volatiles that attract the eulophid egg parasitoid *Chrysonotomyia ruforum,* a species specialized on diprionid eggs (2, 3). We tested whether this response is specific for the plant and herbivore species. *C. ruforum* shows a high specificity for oviposition-induced plant volatiles with respect both to the plant and herbivore species. Furthermore, the parasitoid´s response to these volatiles depends on its experience prior to the bioassay. While numerous generalist parasitoids are well-known to learn a wide range of volatiles when they had the opportunity to associate them positively with a host , the specialist *C. ruforum* was shown just to be able to learn specific volatile blends.

Patch use by egg parasitoids: theory, facts, and biological control

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This is a critical review of theoretical and empirical studies on patch use by egg parasitoids, determining if theory explains observed facts. It explores the possibility that egg parasitoids aggregate in host patches of high density or large size because classical patch-use theory predicts that foraging parasitoids should spend a relatively longer amount of time in high-quality patches, and the possibility that parasitoid aggregation results in density dependent parasitism. It stresses the fact that host age profoundly influences patch use by parasitoids of eggs laid in masses because host quality declines with host age; this occurs simultaneously in all hosts in a patch (i.e. egg mass). When parasitoid females discover low-quality host patches just before host hatching, they should leave the patches as soon as possible to search for another patch. The case of *Anagrus sophiae* (= *delicatus*) females that attack only a small fraction of hosts in an egg mass and leave high-quality patches still carrying apparently mature eggs (Cronin & Strong, 1993) seems to be exceptional among quasi-gregarious egg parasitoids. Characteristics of patch use by egg parasitoids are discussed in relation to their use in biological control.

Selecting arthropod biological control agents: improving agent selection to minimize non-target impacts and maximize target impacts

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Non-target impacts and establishment of ineffective arthropod biological control agents targeting arthropod pests are receiving increased levels of research either through retrospective studies or via *a priori* evaluations before release to from quarantine. Glassy-winged sharpshooter, *Homalodisca coagulata*, an invasive xylophagous hemipteran in California, is the focus of a major classical biological control program with mymarid parasitoids that have been recruited from the pest's home range in the southeast U.S.A. Native sharpshooters potentially at risk from exotic parasitoids are being evaluated for susceptibility to attack as are native sharpshooters in French Polynesia, where *H. coagulata* established *circa* 1999. Laboratory studies assessing risk and parasitoid efficacy have indicated that screening parasitoids for potential efficacy is strongly influenced by parasitoid species and experimental conditions such as the complexity of the environment searched and relative host abundance provide. These different metrics for evaluating parasitoid efficacy can provide different answers as to which parasitoids will be most effective. Our work suggests that greater thought needs to be given to prevailing pest densities at the start of the biological project and how host-parasitoid dynamics will change as successful agents control the target pest making it less common. Lower pest densities because of successful biological control may affect which species or guilds of parasitoids come to dominate and regulate the system at low host densities compared to observations at the start of the program where pest numbers were higher.

Attractants for *Aleochara* spp. (Coleoptera: Staphylinidae), natural enemies of *Delia radicum* (Diptera: Anthomyiidae), in canola

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Defatted mustard seed meal has been shown to attract *Aleochara* species in vegetable brassicas in Sweden, and crop damage was reduced in plots with this treatment (Ahlström-Olsson & Jonasson, I0BC/WPRS Bull. 1992; 15:171). In trials with mustard flour in Canada and with defatted mustard meal in Switzerland, pitfall traps in canola plots with mustard preparations spread at 20 g/m² did not catch higher numbers of *Aleochara bilineata* than did traps in untreated control plots. However, in the Swiss trial, *A. bipustulata* catches in traps in mustard meal plots were almost 2.5 times those in control plots. Parasitism of *Delia radicum* pupae by *A. bipustulata* was significantly higher in mustard meal plots than in control plots, but parasitism by *A. bilineata* was not affected by the treatment. In Y-tube olfactometer studies, *A. bipustulata* exhibited preference for mustard meal compared to a blank control, but *A. bilineata* showed no preference. We are conducting further olfactometer studies comparing mustard meal and mustard flour in the presence and absence of soil and of moisture. Additionally, we are comparing the chemical composition of volatiles from potential attractants using gas chromatograph mass spectrometry.

Influence of host density on the dispersal of a specialist parasitoid

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The use of genetically modified corn in the United States to control the European corn borer, *Ostrinia nubilalis*, is creating a dramatic shift in the spatial dispersion of hosts available to the specialist parasitoid *Macrocentrus grandii*. The persistence of this parasitoid in the Midwestern corn belt may be limited by its dispersal abilities, and its behavioral response to large regions of low host densities created by the widespread use of genetically modified corn. We marked parasitoids developing on *O. nubilalis* using foliar applications of rubidium chloride to 0.2 ha corn plots. Marked parasitoids were presented with transgenic (low host density) and nontransgenic (high host density) corn fields and their dispersal in the two types of fields was monitored using sticky traps. More than 10,000 parasitoids were captured in the first year and preliminary results show differences in the behavior of male and female *M. grandii* dispersing into transgenic corn fields. Parasitoids were captured throughout the 8 ha transgenic corn fields demonstrating that dispersal into low host density fields is possible at this scale. Rubidium levels in parasitoids collected from the 0.2 ha treated plots were 8 times higher than in unmarked parasitoids.

Biological control of *Bemisia tabaci* by the predators

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The sweetpotato whitefly, *Bemisia tabasi* (Gennadius), is a widespread pest of crops in the world, which recently seriously occurs in China, and damages the most economically significant crops of cotton, vegetables and ornamentals, showing a tendency of annual outbreaks with the trouble problem urgent to be solved in agricultural productions. The predatory enemies of *Bemisia tabasi* in China have some species of ladybeetles, thrips, aphids lions and predatory mites, of which the ladybeetles *Serangium japonicum* and *Delphastus catalinae* are active as the dominant species. Both the two species of ladybeetles have a good efficiency feeding on the different stages of *Bemisia tabaci*, especially preferring to the whitefly eggs. At the laboratory, *Delphastus catalinae* develops a generation by 17-22 days at 25-28°C, with the ladybeetble population increases 21.61 times more than that of the fore generation, and develops about 15 generations per year. The predatory quantity per day reaches up 1198 and 670 of whitefly eggs fed by a 4th instar larva and an adult female of *Delphastus catalinae* respectively. The mass rearing of ladybeetles carries out followed by the rearing process of host plant – host whitefly – ladybeetble.

The *Sirex* woodwasp control programme in South Africa

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The *Sirex* woodwasp, *Sirex noctilio*, was first detected in South Africa in the Tokai plantation, Cape Town in 1994. *Sirex noctilio*, together with its symbiotic fungus *Amylostereum areolatum*, inhabits and kills pine trees. Since its initial detection in South Africa, this exotic pest has spread rapidly and is causing increasing losses to the pine industry. Three biological control agents were introduced into South Africa soon after *S. noctilio* was first detected. These included the parasitic nematode, *Beddingia siricidicola*, which is considered to be the best biological control agent against the woodwasp. Unfortunately, the biocontrol agents have not spread together with *S. noctilio*. To address this problem, *B. siricidicola* has been re-introduced on a much larger scale and on a more continuous basis than was previously attempted. The aim here has been to ensure that the nematode becomes established and that it achieves sufficient control of the wasp in South African pine plantations. Studies are also underway to investigate possible differences in the life-cycle of *S. noctilio* in different climatic zones of South Africa. Furthermore, we are considering evidence to suggest that the wasp has been introduced into the country more than once.

Associative learning of plant chemicals by the egg-larval parasitoid, *Ascogaster reticulata* Watanabe (Hymenoptera: Braconidae)

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We examined the learning response to authentic chemicals in *Ascogaster reticulata* Watanabe, an egg-larval parasitoid of the smaller tea tortrix, *Adoxophyes honmai* Yasuda (Lepidoptera: Tortricidae). After 3 times' conditionings of chemicals associated with the host egg mass, the strength of the learned response in a female was evaluated by the distance walked along the chemical cues with her characteristic antennating response in 9-cm Petri dish assays. We compared the learned response of females to 5 authentic chemicals, i.e., linalool, geraniol, methylsalicylate, *(Z)*-3-hexenol and benzyl alcohol. The order of learned responses was as follows: linalool = geraniol > methylsalicylate > *(Z)*-3-hexenol = benzyl alcohol. When the females were conditioned with geraniol, they responded to geraniol significantly longer in a choice between geraniol and linalool. The linalool-conditioned females positively chose linalool in the same choice situation, but not significant. Use of the mixtures of these chemicals in different ratio also had similar effect.

Augmentation of parasitoids by mass-hatching devices: a new biocontrol technique

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Augmentation can be an effective biocontrol strategy to increase the impact of antagonists. However, mass-reared beneficials are generally expensive and often poorly adapted to field conditions. The goal of this study was the development of a device, which selectively permits parasitoids to emerge out of pest-infested plant material. Plant material containing the host and its parasitoids can be collected and stored in large closed devices from which the generally smaller parasitoids can emerge through a selective tissue filter of appropriate mesh size. We used mass-hatching devices to augment populations of indigenous parasitoids of the invasive horse chestnut leafminer, *Cameraria ohridella* (Lepidoptera, Gracillariidae). Whereas more than 75 % of the 15'000 parasitoids passed the separator only 1 % of the 50'000 leafminers overcame the tissue filter. Furthermore, the set-up of mass-hatching devices in the city of Basel, Switzerland, significantly increased parasitism rates of *C. ohridella*. Mass-hatching devices may thus contribute to an integrated control strategy against *C. ohridella* in Europe. In general, mass-hatching devices are a promising new augmentative approach to control insect pests with a dormant stage in or on plant residues and should be considered as a supplementary, cost-effective biocontrol technique.

Molecular genetic investigation of local adaptation and incipient speciation in a tritrophic system

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Seven polymorphic microsatellite loci were assayed in two groups of the insect parasitoid, *Cotesia congregata* (Say) [Hymenoptera: Braconidae]. One group originated from *Manduca sexta* L. and *Manduca quinquemaculata* (Haworth) on tomato and tobacco ("solanaceous group") and the other from *Ceratomia catalpae* (Boisduval) on catalpa ("catalpa group"). Allelic distributions differed significantly between the solanaceous and catalpa groups (Fisher's exact test $P = 0$ overall and for each locus). Across all loci, F_{ST} was < 0 for within-group comparisons and > 0.47 for between-group comparisons. Wasps from the solanaceous and catalpa groups were examined for mtDNA sequence variation to investigate their possible status as sibling species. One conserved mtDNA gene (16S) showed a single fixed nucleotide difference between the two groups. The results support the hypothesis that host plant association is a significant factor influencing population structure of *C. congregata*.

Selecting arthropod biological control agents against arthropod pests: how can the science be improved to decrease the risk of releasing ineffective agents?

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From the practitioner's viewpoint, there are several principles that, if observed, will improve the chances for successful agent selection:

1) Involvement of morphological and/or molecular characterizations is needed from the inception of the project. When the native range/origin of a pest is unknown, this will help to identify significant differences in pest populations and may reveal their actual native range. Identification and characterization of the natural enemy geographic populations and possible host range differences is equally important.
2) Use of climate matching tools is a good starting point for locating climatically-adapted populations of natural enemies.
3) Native range studies should be conducted to evaluate the impact of exotic agents on the target pests.
4) Concurrent and parallel studies should be conducted in the potential release area to document gaps in native enemy activity or other ecological factors that contribute to high pest populations. This also means that a committed practitioner is in place in the receiving area.
5) Collection of natural enemies of species that are taxonomic relatives or ecological homologues of the target pest and which occur in the same or neighboring habitats may reveal field specificities.
6) Quarantine evaluations can provide a preliminary screening of potential effectiveness.
7) Field cage/semi field evaluations are desirable to validate the results of preliminary screenings under more realistic conditions.

These approaches will be discussed in relation to examples of recent projects.

Management of sucking pests in broadacre crops using *Beauveria bassiana* & *Metarhizium anisopliae*.

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Biopesticides based on insect pathogens have been widely applied in an integrated pest management (IPM) strategy against *Helicoverpa* species in Australia, maintaining beneficial insect populations and minimising the development of resistance to chemical insecticides. As reliance on chemical control has reduced in cotton and grain crops, sucking pests such as mirids (*Creontiades* sp.) and green vegetable bug (*Nezara viridula*) are emerging as significant problems. Application of broad-spectrum insecticides against sucking pests threatens the successful IPM strategy by destabilising natural enemy populations and triggering outbreaks of *Helicoverpa sp,* however there are currently no selective insecticides registered for sucking pests. We have identified *Beauveria bassiana* and *Metarhizium anisopliae* isolates active against these pests. Field trials demonstrated control of mirids equal to that achieved with the chemical Dimethoate by four to six days after treatment for both isolates at 1×10^{13} spores/hectare. There was no significant impact on natural enemies in any of the field trials. Monitoring has indicated that sucking pest populations build up in pulse crops through the vegetative stage. We are modelling pest populations and developing a pest management strategy based on population management using biopesticides as an alternative to reactive knockdown using broad-spectrum chemical insecticides.

Red imported fire ant: benefit or detriment to conservation biological control in cotton?

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Predation of sentinel eggs of *Helicoverpa zea* and *Spodoptera exigua* (Noctuidae) was 1.5-2.0 and 4.1 times greater, respectively, in the presence of red imported fire ant, *Solenopsis invicta* (Formicidae), than when fire ants were excluded from 3-ha plots of cotton in central Texas in 2001-2002. Most (68%) of the observed predation events on beet armyworm eggs was due to *S. invicta*. Densities of cotton aphid, *Aphis gossypii*, were 5.5 times greater in the presence of fire ants compared to ant exclusion plots, although aphid densities did not reach economic levels. In additional field trials conducted in 2003, densities of cotton aphid were not significantly different between ant treatments and again did not exceed the economic threshold (50 aphids/leaf). Densities of *Hippodamia convergens* (Coccinellidae) adults and larvae were significantly greater in the ant exclusion treatment Red imported fire ants are intraguild predators in cotton. However, our results suggest that the overall impact of the red imported fire ant is beneficial to cotton pest management in Texas and efforts to enhance fire ant densities have the potential of increasing biological control of nocutid pests.

"Oral toxicity of *Photorhabdus* bacterial toxins against thrips species"

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Photorhabdus and *Xenorhabdus* bacteria are symbiotic to entomopathogenic nematodes. These bacteria are released by nematodes after infection of insects. The oral toxicity of excretion products of several *Photorhabdus* and *Xenorhabdus* strains was tested on two thrips species: *Frankliniella occidentalis* and *Thrips tabaci*. Out of 46 *Photorhabdus* isolates and 6 *Xenorhabdus* isolates only 6 North American *P. temperata* isolates were toxic to the thrips species. After 7 days of inking from *P. temperata* supernatant a mortality of 90% was recorded and reproduction was inhibited. Thrips were also killed after feeding from leaves that were treated with the toxins. Possibilities of using *P. temperata* for the control of thrips will be discussed.

"Understanding target weed population dynamics can improve our ability to select effective agents against weeds"

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Understanding the population dynamics of a weed and the role of environmental factors in influencing these dynamics can help us prioritise insect and pathogen guilds in terms of their likely efficacy in controlling target weeds. In order to be effective, an agent needs to inflict sufficient damage, on appropriate organs or lifestages, and do so in a timely manner. Modelling the population dynamics of a weed can indicate the likelihood of agents from different guilds being able to inflict these desirable damage patterns under the range of environmental conditions that are likely to be encountered in the introduced range. The most desirable agents are those that, when released from their own predators or parasitoids, would act upon the plant population in the manner that most amatically reduces the problems caused by the weed – characteristics that will vary from weed to weed. The characteristics of the most desirable agents can then be used to influence the suite of agents that are specifically targeted for searching, and to help prioritise them for importation and host-specificity testing. If agent establishment follows the lottery model, then prioritising agents that, should they establish, are most likely to be effective should improve our overall success rate.

Efficacy of nuclear polyhedrosis virus with azadirachtin on the mortality, feeding, biological, biochemical & histopathological profiles of *Helicoverpa armigera.*

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The mortality of third instar larvae of *Helicoverpa armigera* larvae was observed in a dose dependent manner and the time to kill was significantly reduced in the combined treatment of Nuclear Polyheosis Virus (NPV) and Azadirachtin (AZA). The average leaf disc consumption of *H. armigera* larva fed with NPV (5 x 10^4 PIB/ml) treatment was 1.90 cm^2/larva and AZA treatment (4 μg/cm^2) was 0.63 cm^2/larva whereas it was significantly reduced to 0.21 cm^2/larva in the low dose combined treatment. Significant reduction in the larval and pupal weights, elongation of larval and pupal duration and a astic reduction in the adult longevity, fecundity and survivability were observed after the combined treatment. Reduced amounts of protein (35.8 mg/g), lipid (47.4 mg/g), carbohydrate (35.7 mg/g), amino acids (32.6 mg/g), DNA at 96 hrs post-treatment (82 μg/g) and RNA at 96 hrs post-treatment (2.4 μg/g) were observed after NPV and AZA treatment. These biochemical changes suggest that AZA did not affect the multiplication of NPV and further there is a synergistic interaction between NPV and azadirachtin. The combined treatment caused a severe necrosis of the midgut epithelial cells and loss of cell shape. The combined treatment of NPV and azadirachtin had much greater effect in relation to the pest management strategy.

Suitability of parasitized greenbugs (*Schizaphis graminum*) as food source for predatory Coccinellidae

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Laboratory studies were conducted to determine first, the stage of development at which the predatory coccinellidae, *Hippodamia convergens*, *Coccinella septempunctata* and C*oleomegilla maculata* are able to start feeding on the parasitized greenbugs "mummies" (*Schizaphis graminum*), and second to determine the nutritional effects of mummies on the growth, development and survival of *Coccinella septempunctata.* Lady beetles were collected from the field and fed unlimited amount of pea aphids (*Acyrthosiphon pisum*) until when they laid eggs. A batch of coccinellid larvae at each instar was fed fresh mummies on daily basis and observed. The preliminary results showed that *Hippodamia convergens* start feeding on mummies at the third instar, even though *Coccinella septempunctata* start feeding on mummies at the second instar, and each instar larva was able to molt to the next stage of development. Further results of the experiment show that *C.septempunctata* take long to develop and they survive through one molt when they are fed mummies. More results will be obtained at a later stage of the study. This study confirms that *Coccinella septempunctata* and *Hippodamia convergens* are able to feed on the parasitized greenbugs at both larval and adult stages of development.

"Isolation & characterization of novel insecticidal *cry1*-type genes from *Bacillus thuringiensis* strains"

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"To identify new *cry1*-type insecticidal genes, 56 lepidopteran-specific *Bacillus thuringiensis* (Bt) strains were isolated from Korea. Universal oligonucleotide primers, K5un2 and K3un2, were designed and used to amplify all known *cry1*-type genes with PCR from 56 Bt isolates. The restriction fragment length polymorphism (RFLP) patterns of the PCR-amplified fragments revealed 7 distinct patterns, and a standard isolate was selected from each one of the RFLP groups. Through cloning and sequence analysis of the PCR-amplified fragments showing unique RFLP pattern, 7 novel *cry1*-type genes were identified. Bt K-1 and Bt 2385-1 strains were selected to isolate and characterize the active regions of the novel genes, and then 5 novel genes were cloned from them. To verify the activity against lepidopteran larvae, the novel genes were expressed using baculovirus expression vector system and the bioassay was performed to *Plutella xylostella* (Px) and *Spodoptera exigua* (Se). All novel *cry1*-type genes had high toxicities against Px larvae and, *cry1-44* and *cry1-5* had the highest toxicity against Px and Se larvae, respectively. Also, *cry1-1* and *cry1-5* had dual toxicities against Px and Se larvae. Finally, each novel gene must be a very useful resource for development of microbial insecticide and insect resistant plants.
"

"Bats feed on crop pests in California's Sacramento Valley"

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Food habits of Mexican free-tailed and Yuma bats were examined in the Sacramento Valley in California. Analyses of guano samples indicated that these two species of bats fed on moths, beetles, and plant bugs suggesting they might help control crop pests. Further studies in cotton, walnuts, pears, and alfalfa showed that bats forage extensively in these crops; however, their impact on biological pest control remains unclear. A colony of 150 bats will feed on more than a million insects a year. In addition, bats may protect crops by their echolocation calls alone. Many insects, including moths, are sensitive to bat calls and take evasive action when they detect bats by turning or diving to the ground, which may disrupt their mating and egg-laying behavior. Given the beneficial nature of bats, growers in California have been putting up bat houses to attract bats to their farms for pest control. Analysis of 175 bat houses in California showed that bats prefer houses that were attached to wooden barns, at least ten feet off the ground, that received morning sun and afternoon shade.

What hypervariable molecular markers tell us about parasitoid population structure & dynamics at both landscape & geographic scales

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We here discuss the use of polymorphic microsatellite markers to study natural populations of the hymenopterous primary parasitoid, *Diaeretiella rapae* (M'Intosh), a polyphagous parasite of crucifer and cereal aphids. Using five such markers, its population structure and dynamics was examined at the field and landscape scale when attacking cabbage aphid, *Brevicoryne brassicae* (L.) infesting oilseed rape, *Brassica napus* at two sites in southern Britain. Parasitoid samples were collected at different spatial scales ranging from individual racemes to field corners. The data were analysed using Bayesian and other statistics to provide new insights into parasitoid movement, persistence and breeding structure. In addition, the same markers were later used to study the genetic variability of *D. rapae* at four geographic sites (Western Australia, France, Morocco and Kazakhstan) reared on the Russian wheat aphid (RWA), *Diuraphis noxia* (Mordvilko). When compared with Old World populations, Australian parasitoids had low allele length and frequency variation, suggesting a significant founder effect. Marked genetic differentiation between populations was also apparent, potentially important for host utilization in this species when introduced to a new geographic area/s. Low genetic variation and gene flow in a founder population could limit evolutionary potential, including the ability to switch to novel introduced hosts, e.g. RWA.

"The density & diversity of parasitic Hymenoptera as bio-indicators of habitat disturbance in a cotton producing region"

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The presence and diversity of hymenopteran parasitoids were compared in cotton with sprays of chemical insecticide, and protected with a bioinsecticide in Huehuetán, Chiapas, México from September, 1999 to January, 2002. Parasitoids were collected weekly from phytophagous insects on upper, middle and lower leaves, squares, flowers and bolls, held in styrofoam cups, with a semi-transparent lid. During the last two years, parasitoids were also collected by vacuuming above the plant canopy. Chemical insecticide decreased both the incidence and diversity of parasitoids early in the first year; however, the guild of natural enemies reappeared and became more abundant and diverse in the chemical treatment. This was associated with the protection brought to the crop by chemical insecticide sprays which resulted in more productive plants, furnishing more refuge to parasitoids late in the season. However, when the crop was rainfed, and phytophagous insects did not cause significant plant damage, natural enemies were affected by the chemical insecticide. Over 300 morphospecies of hymenopteran parasitoids were collected during the experiment. The fact that parasitic Hymenoptera occur at the top of the arthropod food chain makes this group of insects particularly useful in evaluating factors which control and often simplify natural and man-made ecosystems.

The impact of predatory bugs on *Helicoverpa armigera* in Australian cotton crops

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The bigeyed bug, *Geocoris lubra*, and the damsel bug, *Nabis kinbergii*, are two common insect predators in Australian cotton crops. These predators are known to feed on the lepidopteran pest, *Helicoverpa armigera*, however such predation is difficult to observe and quantify under natural conditions. The detection of specific protein markers in predator gut contents allows direct measurement of predation from field-collected individuals. Here, I outline the development of ELISA protocols to detect predation by these two species on *H. armigera* using a species-specific monoclonal antibody. Although prey-specific proteins were detectable for only short periods within the predators, positive examples of predation have been recorded. Adult and juvenile predators of both species were collected from commercial cotton farms in northern NSW during the 2001-02 and 2002-03 seasons. Approximately 12% of *G. lubra* and 24% of *N. kinbergii* tested positive for recent predation on *H. armigera*. Predation rates were similar in both seasons although the relative abundance of *H. armigera* was lower in 2001-02 than 2002-03. This suggests these insect predators can contribute to the suppression of *H. armigera* even when pest populations are low.

Viral pathogens for the control of painted apple moth *Teia anartoides* (Lepidoptera: Lymantriidae)

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The painted apple moth (PAM), *Teia anartoides* (Lepidoptera: Lymantriidae) is a native of Australia, where it is a horticultural pest. Recently, PAM was found in Auckland, New Zealand and poses a threat to horticulture, pine forests and native plants. An eradication campaign was mounted in the Auckland suburbs including aerial applications of the biopesticide Foray (based on *Bacillus thuringiensis*) and releasing sterile male moths. To supplement eradication, we have investigated the availability and potential of microbial insect pathogens to control PAM. Efforts to identify potential biological control agents have included evaluation of several nucleopolyheovirus (NPV) strains from related species (e.g. other lymantriids) and from commonly found forest or horticultural pest insects. Most were non-pathogenic or showed minor sub-lethal effects and one appeared to potentiate a virus present at low levels in PAM larvae. However, an NPV (*Oran*NPV), discovered in PAM in Australia in the 1960s, was highly virulent as were two NPV stains isolated from other lymantriid species (*Orygia pseudotsugata* and *O. leucostigma*). Finally, a cypovirus, isolated from PAM in New Zealand, was infective in bioassays. Preliminary sequence data suggests that this cypovirus is an undescribed member of the *Cypoviridae*. Host range testing of these viruses is underway.

"Occurrence & abundance of spiders in cotton ecosystem of Tamil Nadu, India"

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Agricultural entomologists have acknowledged the importance of spider species as a major factor in regulating pest populations of crops. Despite their importance were highlighted in various ecosystems by several workers, the role they play in regulation of pest populations in cotton ecosystem has received scant attention in India and few workers have attempted on faunistic survey in recording the spider fauna in cotton ecosystem. A survey was conducted at major cotton growing areas of Tamil Nadu *viz.,* Attur, Bhavanisagar and Coimbatore during winter cotton 1998 (July- December) and at Kovilpatty and Srivilliputur during summer cotton 1999 (February – June). The survey yielded 21 species *viz., Araneus* sp., *Argiope catenulata* Doleschall, *A. minuta* Karsh, *Neoscona theisi* Walcknear, *Cyrptophora cicatrosa* Stoliczka, *C. viridanus* Cambridge, *Oxyopes javanus* Thorell, *O. rufisternum* Thorell, *Peucetia viridana* Stoliczka, *Plexippus paykulli* Audouin, *Tetragnatha javana* Thorell, *Thomisus cherapunjens* Tikader, *T. pugilis* Stoliczka, *Leucage decorata* Blackwall, *Gasterocantha citricola* Forskal, *Neoscona elliptica* Tikader, *Clubiona* nr. *assodes* Cambridge, *Lycosa pseudoannulata* Boes. et Str., *Pardosa* sp., *Bianor* sp. and *Salticus* sp. grouped under 16 genera belonging to eight families (Araneidae, Argiopidae, Clubionidae, Oxyopidae, Lycosidae, Salticiade, Tetragnathidae and Thomisidae).

Genetic differentiation of hymenopteran parasitoids: using AFLPs in fingerprinting studies

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"The ecology of insect parasitoids is influenced by plants on which their hosts occur. Parasitoid fitness is affected by plant traits. Thus, adaptation to plant traits could promote significant phenotypic and genotypic differences among sympatric populations. We present data on phenotypic and genotypic differences of two parasitoids ovipositing on (*Plathypena scabra*) on alfalfa and soybean. We compare the developmental time, percent parasitism, and pupal weight of a generalist (*Cotesia marginiventris*) and in a specialist parasitoid (*Aleiodes nolophanae*) and use AFLPs to measure genotypic differences in parasitoids from alfalfa or soybeans. AFLPs have been widely used in biology. Genotypic differentiation among populations of several plant and animal species have been explored with AFLPs. Cluster analyses have been applied to visualize and test systematics, evolutionary and population ecology hypotheses. However, these comparisons are potentially flawed because there is no way of knowing whether the number of individuals and bands used to generate similarity matrices and cluster analyses are sufficient to validly represent the genome of field populations. There are no agreed upon criteria for determining the appropriate number of individuals and bands. We present a method for determining the number of individuals and bands to consider in representing genetic profiles of populations.[agm1]
"

Genetic differentiation of hymenopteran parasitoids

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The ecology of parasitoids is strongly influenced by the host plant. Parasitoid fitness is affected by a variety of plant traits. Thus, it is reasonable to assume that adaptation to plant traits could promote phenotypic and genotypic differences among populations of parasitoids. We present data on phenotypic and genotypic differences of two braconid parasitoids ovipositing on the same host on different host-plants. We compare the developmental time, percent parasitism, and adult weight of a generalist (*Cotesia marginiventris* (Cresson)) and of a specialist parasitoid (*Aleiodes nolophanae* (Ashmead)) of the green cloverworm and use AFLP data as a measure of genotypic differences in parasitoids emerging from hosts on alfalfa versus those reared from hosts on soybeans. AFLPs have been widely used in ecology to explore genetic differentiation among populations in several species and cluster analysis methods have been applied to visualize these differences and to test all sort of hypotheses. However, these analyses are potentially flawed because there is no agreed upon criteria for determining the appropriate number of individuals and bands for such representations to capture variation in the field. We present a method for determining the number of individuals to collect and number of bands to consider when studying genetic differentiation.

Habitat manipulation for insect pest management in Australian cotton cropping systems

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Adoption of within-field monocultures of cotton crops in Australia discriminate against and reduce the establishment and activity of natural enemies of pests because they lack ecological diversity. Increased habitat diversity in the cotton cropping systems has been achieved by planting refuge crops adjacent to cotton fields or as strips or centrally located blocks. The use of refuge crops in this way has enhanced (1) the conservation and establishment of natural enemies on cotton crops early in the cotton season to suppress pest populations and (2) the diversion of pests away from cotton onto the refuge crops resulting in the reduction of pest population. Interplanting cotton crops with refuge crops such as lucerne, the strips should be 8-12 metres wide and 300 metres apart and should run the whole length of the adjacent cotton field. The problem associated with the use of refuge crops for pest management in cotton is that natural enemies may not be inclined to move from the refuge crops to forage the adjacent cotton crops. The movement of the natural enemies can be improved by either applying a natural enemy food attractant to the cotton crops or by reducing the size of the strips planted to refuge crops. The criteria for the selection and management of the refuge crops as well as their deployment in IPM programs in cotton will be discussed.

"Selecting parasitoids for biological control introductions: codling moth as a case study"

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The parasitoid assemblages of lepidopteran pests are more diverse and interactive than those of homopteran pests, posing a greater dilemma for the biological control practitioner in selecting candidates for introduction. How do we identify the single best candidate for introduction, or if more than one species is selected, how important is it that they show no direct interactions? The recent biological control program against codling moth in California will be used to illustrate both theoretical and practical aspects of parasitoid selection for introduction. From a theoretical perspective, the importance of pest refuges from parasitism, antagonistic interactions, and parasitoid aggregation will be emphasized. In contrast, from a practical perspective, the importance of ease of collection, rearing success, and risk will be considered. These contrasting perspectives provide an interesting example of parasitoid selection as both an art and a science.

RESPONSE OF *TRICHOGRAMMA PLATNERI* & *T. PRETIOSUM* TO PATCHES OF SINGLE & CLUSTERED HOST EGGS

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COMMERCIALLY PRODUCED *TRICHOGRAMMA* SPECIES ARE WIDELY USED IN AUGMENTATIVE BIOLOGICAL CONTROL PROGRAMS. HERE WE TEST WHETHER TWO NORTH AMERICAN SPECIES *T. PLATNERI* AND *T. PRETIOSUM*, DIFFER IN THEIR RESPONSE TO AND EFFICIENCY OF USE OF SINGLE AND CLUSTERED HOST EGGS. THE OVIPOSITION BEHAVIOR OF INDIVIDUAL EXPERIENCED PARASITOID FEMALES WAS MONITORED CONTINUOUSLY IN PATCHES (3.5 X 3.5 CM CARD) WITH 16 SINGLE SPACED OR CLUSTERED EGGS OF *EPHESTIA KUEHNIELLA* AT 25°C FOR UP TO 1 H OR UNTIL THE FEMALE LEFT THE PATCH. THE OVIPOSITION BEHAVIOR OF EACH PARASITOID SPECIES IN EACH PATCH TYPE WAS SUMMARIZED AS GENERALIZED ETHOGRAMS, PATTERNS OF PATCH USE, TIME BUDGETS AND THE SEQUENCE OF SEX ALLOCATION. *T. PLATNERI* HAD A GREATER RATE OF OVIPOSITION IN PATCHES WITH SINGLE HOST EGGS, AND AN INDETERMINATE ALLOCATION OF SEXES IN PATCHES OF CLUSTERED HOST EGGS. IN CONTRAST, *T. PRETIOSUM* SHOWED BOTH A GREATER RATE OF OVIPOSITION AND A MORE EFFICIENT SEX ALLOCATION IN PATCHES OF CLUSTERED HOST EGGS. THE MORE EFFICIENT USE OF HOST EGG CLUSTERS BY *T. PRETIOSUM* THAN BY *T. PLATNERI* SUGGESTS THAT CLOSER ATTENTION SHOULD BE PAID TO MATCHING THE OVIPOSITION BEHAVIOR OF *TRICHOGRAMMA* SPECIES TO THE OVIPOSITION STRATEGY OF THE TARGET PEST.

Metarhizium anisopliae as a mycoinsecticide for taro beetles (*Papuana* spp.) in Papua New Guinea & other Pacific Islands.

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Taro is an important root crop for Melanesian and Polynesian people. It is extensively cultivated in PNG as well as Solomon Islands, Fiji, and Vanuatu. The adults of several species of Taro beetle are naturally occuring pests in PNG and the Solomons and recent introductions into other countries such as Fiji. The larvae are detritus feeders and are not an economic problem. The adults burrow down through the soil and eat large holes in the developing corm resulting in the death of the plant or a non-marketable product. An isolate (FI-1452) of *Metarhizium anisopliae* found on *P. woodlarkiana* in PNG is virulent for that species as well as for *P. heubneri* and *P. uninodis*. This isolate is being mass produced on rice in PNG and Fiji and tested in field trials for control of Taro beetle as part of an ACIAR/SPC project. Initial tests in PNG at 3 sites with doses ranging from 10g to 300g applied into the planting hole have increased the proportion of marketable corms from 33% to 65%. Research is still evaluating the most effective dose and an acceptable formulation. Also possible combinations with chemical pesticides are being studied.

Field population dynamics of egg parasitoids of the glassy-winged sharpshooter and smoke-tree sharpshooter in California

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The glassy-winged sharpshooter (GWSS), *Homalodisca coagulata* (Say) (Hemiptera: Cicadellidae), is under chemical management in California in the vicinity of Temecula, the Coachella Valley, the southern San Joaquin Valley, and Ventura Co. This insect is of concern not only because it vectors various strains of *Xylella fastidiosa* with potential to cause disease on a wide variety of crops (e.g., Pierce's disease on grapes) but also because control mandated to limit its spread in the state causes disruption of nursery shipments and movement of harvested citrus. The smoke-tree sharpshooter, *H. liturata* Ball, is native to California and has also been shown to vector *Xylella* diseases although apparently to a letter degree than the introduced *H. coagulata*. A two-year field study in the vicinities of Riverside and Desert Center, CA evaluated levels of egg parasitism on these two sharpshooter species across five fields containing a total of ten host plants. One of these fields was used by the California Deptartment of Food & Agriculture as a site for augmentative release of four exotic species/strains of parasitoids over the duration of this study. The temporal pattern of egg parasitism by eight species, six *Gonatocerus* (Hymenoptera: Mymaridae) and two *Ufens* (Hymenoptera: Trichogrammatidae) are discussed.

Potential biological control of invasive alien flatworms

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The British Isles have been invaded by antipodean terrestrial flatworms, termed locally the 'New Zealand flatworm' (*Arthurdendyus triangulatus*) and the 'Australian flatworm' (*Australoplana sanguinea*). Both species are earthworm predators and there is increasing evidence that *A. triangulatus* negatively affects earthworm populations, in particular anecic species of earthworm. Decline/alteration of earthworm populations could have damaging knock-on effects on soil fertility and other earthworm-feeding wildlife. However, control options are limited due to the flatworms' abundance and cryptic nature. An exclusion experiment was designed to investigate the impact of Carabidae on *A. triangulatus*. Barriered field plots, half of which were treated with insecticide, were sampled weekly for flatworms and carabids. In insecticide-treated plots, there were fewer carabids but more flatworms, compared to untreated controls. However, the difference between treatments was not large, suggesting that carabids moderate rather than control *A. triangulatus* populations. 'Classical' biological control is another option. A Tasmanian midge, *Planarivora insignis* (Mycetophilidae) is the only known parasitoid of terrestrial flatworms. Little is known about this midge beyond a single publication in 1965. Could this midge be used as a control agent for invasive flatworms?

Bollgard$^{®}$II – will it affect abundance of *Microplitis demolitor* in Australian cotton agroecosystems?

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The braconid *Microplitis demolitor* is the most important parasitoid of early instar *Helicoverpa* spp. in the cotton agroecosystem. Late first and second instar larvae are the preferred host stage for oviposition, with host death usually in the third or fourth instar. Feeding by parasitised larvae is greatly reduced, with diet consumption about 10% of that consumed by healthy larvae. In unsprayed non-Bt cotton, parasitism rates of second and third instar larvae often exceed 50% in the latter half of the growing season. *Microplitis demolitor* also transmits ascovirus when ovipositing which usually results in the death of the immature parasitoid in the ascovirus-affected host. Although routine insecticide use in conventional cotton can be disruptive to adults and immature stages of *M. demolitor*, the pupal case offers protection from most insecticides. Cotton is just one of many crop hosts that harbour *M. demolitor* in the farming system. Bt transgenic cotton, particularly Bollgard$^{®}$II, results in extremely high mortality of larvae of *Helicoverpa* spp. It is not known to what extent the increased adoption of Bollgard$^{®}$II will affect abundance of *M. demolitor* through reduced host availability in cotton.

"Quantifying conservation biological control of *Bemisia tabaci* in the cotton system"

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Multiple mortality forces act on insect populations. Estimating mortality caused by natural enemies within the context of other interacting mortality agents is important to understanding the contribution of biological control to pest suppression. Field studies examined the role of natural enemy conservation in cotton within the context of alternative management strategies for *Bemisia tabaci* based on selective and non-selective insecticides. *In situ* life tables were used to quantify and interpret the effects of natural enemies relative to other contemporaneous mortality factors acting on pest populations. Single applications of selective insecticides initially functioned by replacing some mortality from predation and parasitism, and contributing an immediate, and essential, level of irreplaceable mortality. However, because these selective materials did not disrupt natural enemies, parasitoids, and especially predators, were able to supply irreplaceable mortality that contributed to pest suppression for the remainder of the season. This extension in time also served to increase the cumulative chance of pest suppression by weather-related factors. In contrasted systems based on broad-spectrum insecticides, repeated applications were required for pest control. This study demonstrates the role of multiple mortality factors in managing *B. tabaci* and provides a mechanistic understanding of the important contribution of conservation biological control.

"The biology & characterisation of an ascovirus (Ascoviridae) from Australia"

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Ascoviruses (Ascoviridae) are a group of enveloped DNA viruses that cause a chronic and lethal disease in insects. Most of the described ascoviruses are hosted in the larval stage of the Noctuidae (Lepidoptera). Unpublished reports indicated that an ascovirus was found at high incidence (>50%) in populations of *Helicoverpa armigera* (Hübner) and *Helicoverpa punctigera* (Wallengren) in southeast Queensland, Australia. These pathogens were thought to be vectored by the braconid parasitoid wasp *Microplitis demolitor* (Wilkinson). I have formally identified and characterised this ascovirus by examining; the genetic relationship to other known ascoviruses (using RFLPs, Southern blot hybridisations and sequencing the polymerase gene), the host range of the virus and the histopathology. The Australian ascovirus was similar to the *Heliothis virescens* (Fabricius) ascovirus (HvAV) and the *Trichoplusia ni* (Hübner) ascovirus (TnAV) from the USA. To further understand the biology of the Australian ascovirus, I studied its transmission and interaction with hymenopteran parasitoids; I examined other "alternative" modes of transmission and its field prevalence in Australia. The Australian ascovirus appears to be an opportunistic pathogen; it may rely on multiple hosts, multiple vectors and multiple modes of transmission for dissemination and persistence in the field.

Life history of *Euselasia bettina* and *E. chrysippe* (Lepidoptera: Lycaenidae: Riodininae) on *Miconia calvescens* (Melastomataceae) from Costa Rica

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The life history and early stages of *Euselasia bettina* (Hewitson, 1869) and *E. chrysippe* (Bates, 1866) on *Miconia calcvescens* Schrank & Mart. ex DC., are described and illustrated. The species possess six larval instars and the larvae are processionary throughout their development. The eggs are laid in clusters on the underside of leaves—the mean number of eggs in each cluster: *E. bettina* 89 (SD=9; range=82-99; n=3) and *E. chrysippe* 70 (SD=20; range=44-113; n=29). The larvae are gregarious foliage consumers that feed, rest, and molt synchronously. A speices of Aphelinidae was reared from eggs of these two species, and a species of *Calolydella* sp. tachinid fly was reared from late instar larvae of *E. chrysippe*. *M. calvescens* is currently registered as one of the threatening invasive weeds in Tahiti and in Hawaii, and these two *Euselasia* species are currently considered as one of the potential biological control agents (Miconia Project Hawaii-University of Costa Rica 2004). This information will be useful for the biological control workers and personnel responsible identifying insects in quarantine in Hawaii.

"Parasitoid complex of indigenous garden pea leafminer and its effective use for biological control of *Liriomyza* leafminers in vegetable greenhouses"

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The infestation rate of the garden pea leaves by *Chromatomyia horticola* gradually increased during the winter, while parasitoids activity was limited only in a warmer day because of low temperature. However, the parasitoid populations seemed to reach a high density with 90 to 100% total parasitism rate on the well-developed garden pea leafminer populations in late spring and early summer. Throughout 3 years survey, 21 parasitoid species were recorded from the garden pea leafminer, most of which are also parasitoids of the invasive *Liriomyza* leafminers. Among the parasitoids emerging from the garden pea leafminer, *Chrysocharis pentheus* and *Diglyphus isaea* were dominant larval parasitoids, while *Chrysocharis pubicornis* and *Dacnusa nipponica* were dominant pupal and larval pupal parasitoids, respectively. By introducing infested leaves into vegetable greenhouses, the farmer can expect emerged wasps to suppress *Liriomyza* leafminer populations. Changes in the infestation rate by *C. horticola* and its parasitoid complex emphasize the potential in the inoculative biological control of the invasive *Liriomyza* leafminers.

Introducing *Plutella xylostella* granulovirus for DBM control on cabbage through a farmer field school approach in Ghana

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A Kenyan isolate of *Plutella xylostella* granulovirus (*Plxy*GV) was introduced to Ghana for the control of Diamondback moth larvae using a farmer field school approach. Fifty farmers were given training in the use of *Plxy*GV, and in IPM. Field trials were established as part of the training to compare farmers' practice against a control and the use of *Plxy*GV in an IPM context. Density of *P. xylostella* larvae was not significantly different for the three treatments, but pupal density was approximately two times higher on farmer practice plots compared to the control and *Plxy*GV plots. Natural enemies were more abundant on control plots, followed by *Plxy*GV plots and then farmer practice plots. Control of *P. xylostella* larvae alone with *Plxy*GV increased mean cabbage head weight from 0.39 kg for the control to 0.51 kg for the *Plxy*GV treated plants. Mean price per head was US$ 0.2, US$ 0.3 and US$ 0.5 for control, *Plxy*GV treated and farmer practice plots respectively. Reduced phytosanitary intervention frequency on *Plxy*GV plots, reduced cost of control and effective conservation of natural enemies compensated for the relatively lower yields. *Plxy*GV is a safe, effective and environmentally friendly biological control agent and must be popularised in Ghana

Hirsutella thompsonii Fisher, a fungal pathogen of *Floracarus perrepae* Knihinicki and Boczek (Acarina: Eriophyidae)

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The biology and phenology of *Floracarus perrepae* Knihinicki and Boczek, a potential biological control agent of *Lygodium microphyllum* (Cav.) R. Br., Old world climbing fern, was studied in Queensland, Australia from 2000-2002. *Hirsutella* spp. are well-known pathogens of mites and during the study, *Hirsutella thompsonii* Fisher was isolated from *F. perrepae*. The fungus was described and illustrated. Healthy mites are cream in colour but when infected darkened, became moribund and died. Hyphae of *H. thompsonii* filled the abdomen of *F. perrepae* and then protruded from the genital and anal openings and the legs. *Hirsutella thompsonii* caused significant mortality to all life stages of *F. perrepae* in the leaf rolls, causing natural epizootics in field populations.

The successful cross-species release of parthenogenetic *Stichotrema dallatorreaunum* onto New Britian Island, Papua New Guinea, as an IPM agent to suppress tettigoniid pests of oil palm

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Stichotrema dallatorreanum parasitize *Segestidia novaeguineae* (sexava, Tettigoniidae) on mainland Papua New Guinea, but were not known on New Britain Island. Laboratory studies showed that *S. dallatorreanum* from Popondetta reproduce parthenogenetically and could parasitize sexava species found on New Britain (*Segestidia defoliaria* and *Segestes decoratus*). 8431 releases were made on the island during 2000-2002 in oil palm areas to help suppress sexava. Post-release monitoring produced few returns of the parasitoid. Only two recoveries were made in 2002 (near Kimbe and Bialla) and after July no parasitoids were found. In November 2003, a reservoir of the parasitoid was found in *S. defoliaria* near Bialla (>30% parasitization). It is suggested that, because of the severe 2002 dry season caused by El Nino, few hosts were available as sexava eggs were diapausing. In such dry seasons, *S. dallatorreanum* distribution could be reduced to areas with higher rainfall where sexava hatching continues. Similarly, after 1997, *S. dallatorreanum* were only found in wetter areas near Popondetta. To speed up the spread of the parasitoid, parasitized sexava are collected from Bialla and re-distributed into other oil palm areas. However after severe dry seasons *S. dallatorreanum* may have to be redistributed from areas where they have survived.

Performance of *Trichogramma* spp. viewing the biological control of *Spodoptera frugiperda*

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Under laboratory conditions, biological and ethologic aspects of *Trichogramma atopovirilia* and *T. pretiosum* were compared. Under field conditions, the egg-laying distribution of *S. frugiperda* varied according to the phenological stage of the corn. A higher amount of egg laying was collected at 4-6 and 8-10 leaf stages and regardless of the phenological stage of the corn, most egg laying had more than one layer, usually three, covered by scales. The natural parasitism was low and constant in all development stages of the corn and *T. pretiosum* was the most abundant among the collected species. The studies on strain selection showed a wide range regarding performance in *T. pretiosum* strains as to adequacy and preference to *S. frugiperda* eggs. Both *T. atopovirilia* and *T. pretiosum* had a good biological performance and were adapted to *S. frugiperda* eggs even after being reared on *A. kuehmiella* for several generations. The presence of scales on eggs laid by *S. frugiperda* decreased the number of parasitized eggs and affected the behavior of both species. The conclusion is that the parasitism of both species is affected by the physical barriers of *S. frugiperda*, which may lead to a decreased performance of the parasitoid in the field.

Biological suppression of diamondback moth, *Plutella xylostella* (Lepidoptera: Plutellidae) using a cross-infective nucleopolyheovirus of the greater wax moth, *Galleria mellonella*

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A nucleopolyheovirus isolated from the greater wax moth, *Galleria mellonella* (GmNPV) was found to be cross infective to sixteen economically important lepidopteran insect pests, of which seven were confirmed by restriction endonuclease analysis (RENA) of viral DNA. Laboratory bioassays on serial passages of GmNPV five times through seven insect hosts revealed a astic reduction of LC_{50} to the third instar larvae of *Plutella xylostella*. Histopathological studies with GmNPV infected larval tissues of *P. xylostella* showed the rate of viral infection in fat bodies and hypodermis. Field evaluation of GmNPV @ $7.5x10^{13}$ POB/ha against *P. xylostella* on cauliflower indicated that GmNPV reduced larval population by 65.7 per cent, which was next to insecticide, indoxacarb (73.9 %), equivalent to *P. xylostella* granulovirus (65.8 %) and higher than Delfin (59.4 %). The yield of heads (t/ha) also confirmed the above order of efficacy. These studies showed 1) unequivocal confirmation of cross-infectivity of GmNPV to *P. xylostella* by RENA and histopathology 2) possibility of increasing the virulence of virus against targeted pest by serial passages through alternate hosts 3) efficacy of GmNPV against *P. xylostella* under field conditions. The scope of development of GmNPV as a bioinsecticide for ecofriendly management of *P. xylostella* in crucifer ecosystems is discussed.

Importation and establishment of *Bemisia tabaci* parasitoids in southwestern United States

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Bemisia tabaci Biotype B (Homoptera: Aleyrodidae) was first reported causing widespread, economically damaging outbreaks in field crops in southwestern United States in the early 1990's. Shortly thereafter, classical biological control became the centerpiece for a major pest management control program by the USDA and cooperating state agencies. Parasitoids were collected from *Bemisia tabaci* populations in subtropical regions throughout the world. Newly imported, screened natural enemies were released into a combination of urban properties and commercially grown crops infested with *B. tabaci*. Releases of parasitoids imported from overseas began in 1994 and ceased in 2000. Following controlled laboratory and field studies, and small scale field releases, 5 species of Aphelinidae were mass reared and released: *Eretmocerus mundus*, *Er. hayati*, *Er. emiratus*, *Er.* nr. *emiratus*, and *Encarsia sophia*. Differences in species establishment between regions in the United States suggests that pre-adaptation to climate was an important factor to successful colonization.

Using *Trichogramma* to control diamondback moth in cabbage: the influence of alternative host species and plant growth.

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Diamondback moth (DBM) is possibly the most important insect pest in cruciferous crops and there is a global effort to find effective control strategies. Control is only tenuously achieved using insecticides and efforts have long been made to look for alternative control methods. There is interest in the use of the egg parasitoid *Trichogramma* to control DBM. Relatively little work has been done with egg parasitoids and DBM control. Both our laboratory and field studies showed that *Trichogramma* could definitely be an important component of an IPM control program of DBM. The *Trichogramma* species used in the study was shown to prefer the smaller DBM eggs to the larger eggs of the soybean looper, an alternative host species. Prior exposure to one or other of the host species' eggs did not affect this preference. The fieldwork showed that levels of parasitism greatly depended on the interactions of cabbage plant growth, ovipositional patterns of the host species and foraging behavior of the *Trichogramma*. This study highlights the fact that successful use of *Trichogramma* (and other biological control agents) depends on piecing together the pertinent interactions of host, host plant and parasitoid, ensuring correct and effective use of the biological control agent.

Bio-efficacy of *Heterorhabditis indicus* (RCR) along with three botanicals against *Helicoverpa armigera (Hub.)*

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Compatibility of locally isolated entomopathogenic nematode, *Heterorhabditis indicus* (RCR) in different concentrations of leaf extracts of *Prosopus juliflora, Vitex nigundo* and *Pongamia pinnata* and their bio-efficacy against *Helicoverpa armigera* (Hub.) was conducted under laboratory condition during 2002-03. In compatibility study, all botanicals tested were highly toxic to infective juveniles (IJs) at higher concentrations (10, 5 and 4 %) resulting in more than 80 % mortality after 48 h of exposure. As the concentration reduced, the nematode survivability increased. Lowest concentration of one percent resulted 34.83, 30.10 and 31.2 % nematode mortality in *P. juliflora, V. nigundo* and *P. pinnata*, respectively indicating the scope of integration. In bio-efficacy study, *H. indicus* at 50 IJs/larva in combination with 0.5 % of *P. juliflora* and *V. nigundo* recorded cent percent mortality in early third instar larvae after 48 h and was on par with inoculation of nematode alone at 150 IJs/larva. However, similar combination in *P. pinnata* resulted in only 65 % mortality. Sequential application of either *P. juliflora* or *V. nigundo* at 2.5 % followed by nematode at 50 IJs/larva was the next best option which resulted in 70% mortality. Similar trend was observed against late fourth instar larvae.

Movement of the predatory coccinellid, *Hippodamia convergens*, in a two-crop system

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Complementary field studies were used to investigate predator movement between grain sorghum and cotton crops. Rubidium mark-capture experiments suggested that coccinellids, particularly *Hippodamia convergens* Guérin-Méneville, produced an overall pattern of predator movement from sorghum into cotton. Manipulations of aphid levels in field cages tested if prey abundance and crop phenology influenced movement of *H. convergens* into cotton. More beetles were collected on caged cotton during late stages of crop phenology or when relatively low cotton aphid densities were present. *H. convergens* carbon isotope ratios were used to assess adult feeding after colonizing cotton and whether prey consumed in sorghum contributed to egg production in cotton. When cotton aphids were absent, *H. convergens* adults stayed in cotton, did not produce eggs and consumed few prey. With cotton aphids present, *H. convergens* isotope ratios changed from prey consumed in cotton, and isotope ratios of egg masses indicated prey consumed in grain sorghum contributed very little to egg production in cotton. An area-wide pattern analysis also suggested the abundance of grain sorghum and uncultivated areas positively influenced cotton predator abundance, as did prey levels and cotton planting dates.

"Management of insect pests of cole crops through utilization of biotic agents in northwestern Himalayas, India"

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The studies were conducted for three consecutive years in three districts of ecological safe hill state where pesticidal applications have recently started picking up on vegetables. In view of giving pesticide free cabbage, cauliflower and broccoli, field releases of biotic agents viz., *Trichogramma chilonis* and *Trichogramma bactrae* @ 75000/ha against lepidopterous pests were made at all the locations in three districts *viz.,* Kangra, Mandi and Una during cropping seasons in 1998,1999 and 2000.*Bacillus thuringiensis* was also applied @ 2.5 g/litre of water.*Diaeretiella rapae* was released @ 50000/ha against aphids.There was considerable reduction in the population of *Pieris brassicae* and *Plusia orichalcea* when Bt sprays were given along with parasitoid release. However, good control of *Plutella xylostella* and *Helicoverpa armigera* was achieved even without Bt spray with the releases of *T. bactrae* and *T. chilonis*. Bt sprays further added to achieve better control. The population of aphids generally remained low. But the releases of *D. rapae* led to more decline in aphid population

Insecticide use pattern and locational specific performance of *Cotesia plutellae* Kurdjumov in parasitization of *Plutella xylostella* L. larvae

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Insecticide use pattern and parasitism of diamondback moth (DBM), *Plutella xylostella* L. by *Cotesia plutellae* Kurdjumov was surveyed under field conditions in North East Plain regions of India. Farmers used only the insecticides to combat DBM and the insecticide use pattern against DBM during the two years of study revealed that organophosphates were the most commonly used insecticides (30.99 % and 44.87 %) followed by synthetic pyrethroids (30.99 % and 23.08 %), chlorinated hydrocarbons (22.53 % and 15.38 %), carbamate compounds (14.08 % and 12.82 %) and botanicals (01.41 % and 03.85 %) during the first and second years of the experimental periods, respectively. Among various chemicals, cypermethrin was most extensively used (26.76 % and 21.79 %) followed by cartap hydrochloride (14.08 % and 12.82 %), endosulfan (12.68 % and 11.54 %), quinalphos (11.27 % and 07.69 %) and dimethoate (05.63 % and 12.82 %) during the two years of experimentation on cauliflower. The *C. plutellae* pupae, the only abundant larval parasitoid of DBM in this region were collected from these different locations having varying insecticide pressure. The killing efficiency (K-value) and the longevity of the female parasitoid collected from the areas of higher insecticide input was more (0.805 and 0.790) than those collected from the areas receiving less insecticides (0.706 and 0.577) during both the years of study.

Coenosia humilis an important predatory muscid of *Liriomyza huidobrensis* attacking potatoes in Indonesia

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Field and laboratory studies were conducted on potatoes in Pangalengan, West Java to (a) determine the abundance of the predatory fly *Coenosia humilis* Meigen (Diptera: Muscidae) and the potato leafminer fly *Liriomyza huidobrensis* (Blanchard) (Diptera: Agromyzidae). (b) study the behaviour of the predator and (c) evaluate effects of insecticide application on both flies. Our research revealed that the abundance of *C. humilis* and *L. huidobrensis* both peaked 56 days after planting of the potato crop. Yellow sticky trap catches showed that adults of *C. humilis* and *L. huidobrensis* were both more active during morning and afternoon than in the middle of the day. In the field, some 113 *C. humilis* flies were collected with prey sticking on their mouth parts. Laboratory examination indicated that the prey consisted of small sized insects such as Diptera and Homoptera, with the highest proportion (68%) being agromyzid flies. In the laboratory, each predator consumed an average of 23.5 *L. huidobrensis* flies in 24 hours or about one fly per hour. Under field conditions, predation rate might be lower. However, since the predators were abundant in potato fields in Pangalengan, *C. humilis* might be expected to make a significant contribution in suppressing leafminer fly population. Insecticide applications as practiced by farmers significantly suppressed the predator and at the same time promoted the leafminer population.

Antioxidants increase the efficacy of commercial baculovirus biopesticides against *Helicoverpa* sp. in cotton

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Commercial biopesticides based on nucleopolyheoviruses (NPVs) are widely used to control *Helicoverpa* sp. in eastern Australia. While results are generally good, the virus is less effective on cotton than on other crops. One hypothesis for this is that the surface of the cotton leaf is hostile to baculoviruses, resulting in rapid loss of persistence. Another hypothesis is that constituents of cotton foliage interfere with the infection process in the insect midgut. Hoover *et al* suggested that this is due to oxidation by free radicals generated during consumption of the leaf, and might be modified by applying antioxidants such as mannitol with the virus. We tested this hypothesis in the field by applying NPV with mannitol, which resulted in a 50-75% increase in insect mortality. Laboratory experiments indicated that mannitol is not a feeding stimulant, nor did it activate the virus directly, which supports the hypothesis that the increase in mortality is due to interactions of mannitol with the cotton plant chemistry. Mannitol might serve as a useful additive to NPV products. However, it is relatively expensive and would significantly increase costs to growers. We tested alternative compounds that delivered similar increases in efficacy at reduced cost.

"Beating around the bush in Australia: native vegetation resources for beneficial insects"

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"A range of beneficial insects species occur within the cotton agroecosystems in Australia, but farming systems do not always encourage their survival. Recent changes in cotton farming systems and an increased awareness of the value of pest control services provided by generalist predators have led to increased enthusiasm for maintaining populations of beneficial insects on farm. Some efforts have been made to plant winter wheat, sorghum and lucerne to increase in-situ populations of beneficial insects. Little attention, however, has been given to the existing non-crop vegetation (native trees, grasses and shrubs and introduced weeds) surrounding cotton fields and the role it may play in supporting populations of beneficial insects. The aim of this study is to investigate the spatial and seasonal fluxes of populations of several species of generalist insect species within the surrounding non-crop vegetation, thus learning more about potential habitats for beneficial insects within the cotton agroecosystem and finding ways to preserve and incorporate these habitats within existing cotton farms.
"

Biotechnology and maize pests: historical farmer perceptions and adoption of transgenic maize in integrated production

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Ostrinia nubilalis (Crambidae), *Diabrotica virgifera* and *Diabrotica barberi* (Chrysomelidae) are primary maize pests in the United States. Transgenic maize expressing a *Bacillus thuringiensis* (Bt) protein for *O. nubilalis* control was commercialized in 1996. Farmers (n=7,427) that planted Bt maize were surveyed (1996-1998). Farmers planting Bt maize increased acreage from 10.5% (1996) to 40.7% (1998) and decreased insecticide use from 13.2% (1996) to 26.0% (1998). The primary reason for planting Bt maize was to eliminate insect-caused yield loss. Most farmers believed Bt maize yields were similar to or greater than non-Bt hybrids and that *O. nubilalis* previously caused greater yield loss than suspected in non-Bt maize. A second survey (2001) evaluated farmer (n=1,313) perceptions prior to commercialization of transgenic Bt maize designed to control *Diabrotica* spp. Most farmers (75.2%) used crop rotation to control *Diabrotica* larvae. However, 33-39% of farmers in two major maize states used insecticides in rotated maize. Perceived benefits of *Diabrotica*-control maize were less insecticide in the environment (68.5%) and less insecticide exposure (69.9%). Concerns were selling the grain (59.3%) and higher technology fees (54.8%). These surveys provide an historical foundation on transgenic Bt maize use, initial farmer perceptions regarding performance, and attitudes regarding pest control.

Genetic, molecular, and analytical tools for refining classical biological control

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The field of population biology has fully embraced the use of novel genetic and molecular tools and accompanying analytical approaches to understand both population dynamics and micro-evolutionary change. Here, we attempt to demonstrate how classical biological control can benefit from many of these approaches, and in particular, to address the charge that "biological control is not yet a predictive science." First, we examine the following processes: agent selection/prioritization, diagnostics and identification, quarantine, efficacy, and target specificity. For each we point to studies in the basic literature that have used successfully molecular, genetic, and analytical approaches to address similar issues. Second, we argue that classical biological control may be more predictive than recognized, especially when biological control is considered as replicated semi-natural field experiments. In this light, the field has much to offer as a test bed for theories in both ecology and evolutionary biology. Lessons from biological control can even help inform researchers and policy makers about the risks of both invasive organisms and genetically modified organisms.

Impact of insect-resistant transgenic crops on entomophagous arthropods and how to assess these effects

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"One of the primary concerns related to the adoption of insect-resistant transgenic plants is their potential impact on non-target organisms. This includes entomophagous arthropods which play an important role in natural pest regulation. Despite the fact that regulatory bodies require information on the potential risk of releasing transgenic plants in the environment, to date, no specific protocols have been designed for assessing the risks that insect-resistant transgenic crops pose for these beneficial organisms. Here a framework for risk assessment is proposed to evaluate the effects of insect-resistant plants on entomophagous arthropods. Using maize expressing the *cry1Ab* gene derived from *Bacillus thuringiensis* (*Bt*) as an example, we illustrate the steps necessary for assessing the risks for larvae of the green lacewing, *Chrysoperla carnea*. Based on the information available we conclude that the *Bt*-transgenic maize poses a negligible risk for this important predator. The results are presented in the context of other laboratory studies on the impact of *Bt*-transgenic crops on entomophagous arthropods.
"

Bt-transgenic cotton and insect natural enemies: compatibility or irreconcilable differences?

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Transgenic insecticidal plants confront herbivores and their natural enemies with a potent bottom-up force. Targeted herbivores are directly and severely affected by the transgene product, whereas nontargeted herbivores may experience direct or indirect consequences from reduced competition or modified pest management practices. Our studies focused on effects of Bt-cotton on population dynamics of predators in fields in southern Georgia, USA. We examined a suite of foliage-dwelling and epigeal predators in cotton fields (paired Bt- and non-Bt cotton fields). We found no differences in abundance or diversity of predators over 2 years. We also assayed selected species of field-collected heteropteran predators for presence of Bt protein. Most predators contained no detectable levels of Bt; some nabids did. We also examined direct and indirect effects of Bt-cotton on the predator *Geocoris punctipes* by rearing bugs on Bt- and non-Bt cotton with two prey: 1) eggs of *Helicoverpa zea*; and 2) 1-2 day-old larvae of *Spodoptera exigua*. Predator development, size, fecundity, and longevity were affected by prey type, but no plant effect was observed, indicating that Bt-cotton does not adversely affect this omnivore directly or indirectly. To date we have found no detrimental effect on predator populations in the cotton agroecosystems.

"Genetic polymorphism in prey preference of a polyphagous predatory arthropod"

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Arthropod predators usually feed on a variety of prey types. At one extreme, populations consist of an ensemble of specialists and at the other they are composed of indiscriminate generalists. In between these extremes populations may be composed of generalist predators with learned or genetically fixed prey preferences. Whether mono- or polymorphism will evolve depends on various factors, such as the cost of flexibility for the generalist, the variability in supply of various prey species and the degree to which the population is homogeneously mixed with respect to mating. Here, we report on experimental evidence for genetic variation in a local population of the haplo-diploid predatory mite, *Hypoaspis aculeifer*, collected from *c.* 0.25 m^2 sandy soil in a lily field. Using two species of astigmatic mites as prey (*Rhizoglyphus robini* and *Tyrophagus putrescentiae*) two-way selection for prey preference during four generations yielded lines with preference for one or the other prey. Cross-breeding in both directions resulted in hybrid females with intermediate preferences and the choices found in females from F1xParent backcrosses were best explained by assuming monogenic inheritance without dominance. We also demonstrate that this genetic polymorphism can be maintained by heterozygote advantage and disassortative mating.

Oligidic diet on the biology of a reduviid predator *Rhynocoris marginatus* (Fab.) (Heteroptera: Reduviidae)

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Rhynocoris marginatus (Fab.) (Heteroptera : Reduviidae) belongs to Harpactorinae a sub family of Reduviidae. It has been reported to predating upon more than 20 insect pests belongs to various orders. However, mass production technique is not available for this promising biological control agent. In this work we tried to record the biology of this reduviid on beef liver based oligidic diet (OD). It revealed that all the life stages of *Rhynocoris marginatus* preferred to feed the oligidic diet, but the preference varied depends upon the nymphal stage. For instance, first nymphal instar highly preferred the OD (74.5%) followed by fourth nymphal instar (70.35%). Oligidic diet prolongs the nymphal developmental period from 44.21 days (reared on factitious host *Corcyra cephalonica* Stainton) to 123.0 days. Similarly oligidic diet reduced the survival rate and weight gain. The weights of the adult *Rhynocoris marginatus* were 68.33 and 107.0 grams when reared on oligidic diet and *C. cephalonica*. Oigidic diet also alters the sex ratio (0.75 and 0.58 for OD and *C. cephalonica,* respectively). Oligidic diet reared *Rhynocoris marginatus* do not laid any eggs during its adult stage. We propose to alter the adult diets for the successful mass production of this reduviid.

Microbial control of the red palm weevil *Rhynchophorus ferrugineus* (Oliv.) in Egypt

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**"The disastrous invasion of the red palm weevil *Rhynchophorus ferrugineus* results in great losses of palm trees ever encountered in Arab Gulf countries and is entitled as the palm tree acquired immune deficiency syndrome (AIDS). The female is a strong flyer and lays its eggs in wounds around the trunk of palm trees. The insect development from hatched larvae to adult is completed entirely inside the trunk of the tree causing damage by feeding on tissue that may ultimately lead to death of tree. In this microhabitat, the conditions are more favourable than the outer atmospheric conditions. The cytoplasmic polyheosis virus was isolated from the larvae, identified and its symptomatology was considered. Based on laboratory studies, large scale field experiments were conducted to investigate its pathogenicity when injected inside the trunk of the infested trees at a concentration of 100×10^6 polyhedra / ml at the rate of 200 ml / tree . After 3 – 4 weeks, complete recovery of infested trees occurred . The mirosporidia *Vairimorpha ephstiae* showed also to be highly potent when injected at a concentration of 100×10^6 spore / ml at the rate of 200 ml / tree . A complete recovery of the trees was obtained within 4 weeks. Combination of both agents led to high synergism Both agents or their combination may be recommended for control of this curculionid insect pest most efficiently .
"**

Mass rearing methods for *Metaphycus bartletti* and *M. luteolus*, biological control agents of mediterranean black scale and soft brown scale

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Mass rearing techniques of the mediterranean black scale, *Saissetia oleae* and soft brown scale, *Coccus hesperidum* , hosts for parasitoids *Metaphycus bartletti* and *M. luteolus* were tested under laboratoyry conditions at 24.4±2°C, 55±5% RH and L:D 14:10 in Biological Services in Loxton, South Australia. Black nightshade, *Solanum nigrum* was among the best plants with more than 76 and 80% crawlers establishment for black scale and soft brown scale, respectively. Second and third instars nymphs were transferred to butternut pumpkins, *Cucurbita moschata* for adult scales reproduction. The mean of one generation of black scale and soft brown scale was completed in 73.9±1.8 days and 44.4±2.4 days, respectively. The best black scale stage for *M. bartletti* production were 3rd and 4th instars and for *M. luteolus* were 2nd and 3rd instars. Both female parasitoids did not attack 1st instar and mature adult scales. Female *M. bartletti* parasitised proportionally more hosts as host density increased meanwhile, low percentage of parasitism by female *M. luteolus* was estimated by increasing number of soft brown scale. Results indicated that parasitized scales could be stored in the refrigerator at 10°C at least to 8 weeks without any effect on adult parasitoids.

The use of rubidium as a marker to assess the dispersal of the parasitoid, *Dolichogenidea tasmanica* (Hymenoptera: Braconidae) in vineyards

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Dispersal of the parasitoid, *Dolichogenidea tasmanica* (Cameron) (Hymenoptera: Braconidae) which parasitises larvae of the lightbrown apple moth, *Epiphyas postvittana* Walker (Lepidoptera: Tortricidae) was studied from flowering buckwheat, *Fagopyrum esculentum* Moench plants in a vineyard in New Zealand. Four sequential foliar applications of rubidium chloride (RbCl) were applied to single strips of buckwheat, sown in the centre of each of five vineyard blocks. Yellow sticky traps were placed in each block at distances of 0, 2, 5 and 15 vine rows from the buckwheat to collect adult *D. tasmanica.* Buckwheat plant tissue samples were taken following the application of RbCl to ascertain whether the plants had been marked. Results showed that the number of *D. tasmanica* marked with rubidium decreased linearly from the buckwheat. Also, the number of *D. tasmanica* caught was significantly higher at distances of 0, 2 and 5 vine rows from the buckwheat. The buckwheat plant tissue was marked with rubidium and these levels declined after the initial application of rubidium. These results suggest that *D. tasmanica* fed on the buckwheat plants and then moved at least a distance of 10 meters from the plants within the seven-day sampling period.

Landscape and on-farm dispersal of *Diadegma semiclausum*: a key parasitoid of the diamondback moth, *Plutella xylostella*

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On-farm refuges can be effective for conservation biological control if beneficial insects move from the refuge to the crop in greater numbers than the pests. We conducted experiments to monitor on-farm and property-property dispersal of the parasitoid, *Diadegma semiclausum* (Hellén) (Hymenoptera: Ichneumonidae). We sprayed a resin-based fluorescent dye on the refuge crop to mark the resident insect populations and monitor their dispersal before and after the refuge was cultivated. Before cultivation of the refuge, 5% of marked *D. semiclausum* had dispersed as far as 60m. After cultivation, 7% had dispersed as far as 212m (the furthest distance sampled), while greater than 85% of *P. xylostella* (L.) (Lepidoptera: Plutellidae) moths moved to the adjacent broccoli. Within a crop, dispersal of marked *D. semiclausum* was slightly aggregated before cultivation, but randomly distributed after cultivation. Among farms, at the scale of the landscape, 10-17% of *D. semiclausum* moved between properties greater than 1.2 km apart, and they were highly aggregated in brassica crops. *D. semiclausum*, and *P. xylostella* were highly associated at the landscape scale. Our results suggest that on-farm refuges provide a source of *D. semiclausum* that move into adjacent plantings, and between properties.

The establishment and utilisation of *Trichogramma pretiosum* Riley against *Helicoverpa* spp. in Australian cotton farmscapes

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Trichogramma pretiosum is a New World species of egg parasitoid that has been evaluated and utilised as a biological control agent of *Helicoverpa* and *Heliothis* pest species throughout the world. It was released in the Ord River irrigation area of Western Australia, and on the Darling Downs region of south-east Queensland, during the 1970's. The species became established in the Ord River region, but was not recorded during extensive sampling of *Helicoverpa* eggs on the Darling Downs from 1985-1995. Releases of *T. pretiosum* recommenced on the Darling Downs in 1995, and the species has since become widespread and abundant in cotton farming systems. *T. pretiosum* now accounts for 97% of all egg parasitoids reared from *Helicoverpa* eggs in cotton on the Darling Downs, and has caused high levels of egg parasitism (80-100%) from mid-January to April for the past six years. The introduction of *B.t.* cotton, and greater use of selective insecticides, are thought to have benefited *T. pretiosum* by reducing the number of disruptive broad-spectrum chemicals used to manage cotton pests. The diversity of cropping on the Downs, and the continuity of hosts that this favours, is also believed to assist the growth of *T. pretiosum* populations.

"Enhancing juvenile mortality of *Helicoverpa* spp. (Lepidoptera: Noctuidae) in chickpea/cotton cropping systems"

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This presentation is a summary of research aimed at finding non-chemically dependent ways of minimising the survival of *Helicoverpa armigera* and *H. punctigera* in chickpea (*Cicer arietinum*). Chickpea is highly attractive to *Helicoverpa* in all its phenological stages. Field assessments of egg distribution patterns reveal marked changes in the oviposition behaviour of *Helicoverpa* spp. in chickpea plots with and without contaminant plants of other species. Comparisons of mean egg densities on contaminant plants and the surrounding chickpea crop indicate that moths lay most of their eggs on the contaminants. Characteristics of this "aggregative" egg distribution phenomenon are described in relation to crop phenology within contaminated crops. Life table assessments indicate that > 80% of eggs laid on the contaminants do not survive to the larval stage. Our results suggest that contaminant plants in chickpea plots serve as *Helicoverpa* sinks. We propose that differential host plant apparency is a plausible explanation for the aggregative oviposition response to and the resultant sink effect of contaminants in chickpea. A strategy of deliberate contamination of chickpea crops with other plant species merits further investigation as a cultural pest management strategy for *Helicoverpa* spp. Implications of *Helicoverpa* management in chickpea for cotton pest management are discussed.

"Influence of radiation on parasitoid - host interaction between *Steinernema glaseri* and *Spodoptera litura* (Fab.) vis-à-vis other biorational pest control measures"

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"Bioinfectivity of entomopathogenic nematodes (EPNs) was evaluated on radio-sterilized host, followed by assessment on perpetuating bioefficacy of EPNs derived from irradiated hosts, up to ensuing two generations, to ensure safe transport and viable augmentation of this bio-control agent. Further, the interaction of EPNs was studied with radiation-induced F_1 sterility in *S. litura.*, by evaluating the infective behaviour of normal infective juveniles (IJs) on F_1 sterile insects, and viability of F_1 IJs derived from F_1 sterile host, in order to understand the feasibility of using EPNs with F_1 sterility technique for pest control. Behaviour of EPNs from radio-sterilized hosts in relation to host treated with moulting hormone (MH)-agonist was studied. Both the sterilizing and lethal doses of MH-agonist, appeared to be compatible for integrated application with bioinfective EPNs. Also, the effect of low dose ionizing radiation on EPNs bio-efficacy was studied towards host in different regimens, in order to explore radiation hormesis. Low dose range (1.5-2.5Gy) could induce faster morbidity and mortality in host by irradiated EPNs, with their slightly enhanced harvesting, when irradiated IJs were bioassayed one week post-irradiation. Potential role of radiation as a tool in pursuing compatible utilization of these EPNs with biological based tactics is discussed.
"

Novel methods of entomopathogenic nematode application

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Entomopathogenic nematodes are generally applied for insect control in aqueous suspension using various sprayers or irrigations systems. Novel methods of application can reduce costs and improve efficacy of pest suppression. Some novel methods of application include baits or slow-release bags. Another alternative is application of nematodes in their infected hosts. Applications of nematodes in infected hosts can result in significant pest suppression under field conditions. Our research has addressed the following questions: 1) Are there any advantages to applying nematodes in infected hosts Vs aqueous application? 2) Is it feasible to apply infected hosts from a technical standpoint? Our laboratory experiments suggest advantages of greater dispersal, infectivity, and survival in nematodes applied in infected hosts compared with aqueous application. In greenhouse studies, superior efficacy was observed in suppression of *Diaprepes abbreviatus* and *Otiorhynchus sulcatus* when nematodes were applied in infected hosts. Further, our studies indicate it is feasible to store and package nematode infected hosts for commercial application. Fragile cadavers such as *Galleria mellonella* can be coated with formulations to facilitate storage and application. Alternatively, hard-bodied insects (such as *Tenebrio molitor*) can be ideal hosts because they are naturally resistant to rupturing or sticking together.

Molecular characterisation of the whitetop collar gall weevil, *Ceutorhynchus assimilis,* opens the door to its use as a biological control agent

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Molecular phylogenetic studies have increasing relevance in biological control for (a) subspecific genetic characterisation of the target to trace origins or relatedness to non-targets, (b) the subsequent design of host test lists for risk analysis, and (c) subspecific genetic identifiers of agents to allow monitoring following release. A recent exciting development has used of molecular techniques to create biocontrol opportunities for potential agents previously confounded by taxonomic ambiguities or complex subspecific host specificity variation. In this paper we overview one such study that mapped population level mitochondrial haplotypic variation of the collar gall weevil, *Ceutorhynchus assimilis* (a natural enemy of the white-top, *Lepidium aba*, an exotic invasive weed across 3 continents) to differences in host specificity. Literature and field records suggested the weevil had a relatively broad host range within the Brassicaceae. The biogeographical distribution and field host range of 4 morphologically indistinguishable clades of the weevil were defined from geographically widespread collections. Only the target weed supported complete larval development in one of these clades. A parallel origin-tracing study evaluating the genetic structure of *Lepidium aba* in its native range can now be used to assess the non-target risks from inferred insect-plant evolutionary patterns.

Economics of transgenic management programs: what are the global implications of IPM community acceptance?

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US growers are planting 80 million acres with transgenic crops because the crops are the most cost-effective means for control of certain pests. The transgenic crops are less expensive and/or more efficacious in controlling the pests than are conventional crops. US growers are gaining $1.5 billion in annual net income from planting transgenic maize, soybeans, cotton, canola, squash and papaya. Transgenic crops in development in the US for pest management could add another $1.0 billion to annual net income. Potential economic gains of planting transgenic crops are also very high in the European Union. Transgenic herbicide-tolerant oilseed rape, maize, rice, wheat and sugarbeet are projected to improve EU farm net income by $561 million while fungal resistant potato and insect resistant maize would provide approximately $1.2 billion in net benefits. When transgenic crops are used to manage pests, one result is a reduction in the use of chemical pesticides. In the US, transgenic insect-resistant maize and cotton substitute for 2.6 and 1.9 million pounds of insecticides. In the EU, a fungal-resistant potato would eliminate the need for 7.5 million kilograms of fungicides.

How can understanding target pest and agent population dynamics improve our ability to select effective agents against arthropod pests?

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First we reviewed the contents of several entomological/biocontrol journals to see where biocontrol research is focused and in what directions researchers think it is headed. Second, we used meta-analysis to look at pest densities in the presence and absence of biological control agents. Various biological features of both pest and natural enemy were coded in the analysis to help us understand which biological features are important in reducing pest densities and thus increasing the likelihood of selecting effective biocontrol agents.

"Evaluation of *Beauveria bassiana* for control of household ants"

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"Efficacy of the entomopathogenic fungus, *Beauveria bassiana*, was evaluated for household ants in indoor environments. Efficacy was determined by exposure of ants to baits or baited traps. Five ant species (fire ant, carpenter ant, pharaoh ant, crazy ant and ghost ant) were exposed to Fungus biological bait (*B. bassiana*) or chemical bait from commercial products (Combat, Max, Raid) for 3 days. Also, fire ants were continuously exposed for 90 days to fungal bait powder in plastic traps. Efficacy was characterized by cumulative percent mortality. At 28 days after initial exposure to baits, mean cumulative mortality of ants ranged from 26%-100%. No bait gave the highest mortality for all ant species but Max was the most efficacious for 4 species, Fungus and Combat for 3 species and Raid was not best for any species. Fungus had the highest weighted sum score for all ant species tested. Death was delayed when fungal powder bait was placed in plastic traps and ants had to search to find bait. When fire ants foraged bait in traps, mortality did not reach 90% until 70 days of continuous exposure.
"

Potential benefit of the fire ant – cotton aphid mutualism for cotton plant fitness

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Ants commonly form mutualistic associations with honeydew-producing homopterans in which homopterans gain protection from predators and parasitoids in return for producing a food resource (honeydew) for ants. The cost to plant fitness of this mutualism can be substantial and represents a real threat to yield in agricultural systems. Plants may benefit from the mutualism, however, if the attraction of ants onto plants increases ant suppression of other, non-homopteran herbivores. Red imported fire ants (*Solenopsis invicta*), an abundant introduced predator, engage in a mutualism with cotton aphids (*Aphis gossypii*), an important pest of cotton, throughout the southeastern United States. We hypothesize that low to moderate densities of cotton aphids indirectly benefit cotton yield by attracting fire ants onto plants resulting in increased suppression of caterpillar herbivory. Results from field experiments are consistent with our hypothesis: fire ant abundance on plants increased significantly with cotton aphid abundance, resulting in decreased plant damage by caterpillars and increased square and boll number. Our results highlight the potential indirect benefit of homopteran pests in biological control, but we argue that the benefit is conditional. We develop a heuristic model that predicts that the benefit of the fire ant-cotton aphid mutualism varies with the intensity of caterpillar herbivory.

Diversity of parasitoid fauna of *Liriomyza* Spp. (Diptera: Agromyzidae) on vegetable crops in Bali and Lombok

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The detailed surveys for parasitoids of leafminers in Bali and Lombok were conducted from June 2001 until October 2003. There were 14 parasitoids associated with *Liriomyza* spp. on vegetable crops in Bali and Lombok. Among the 14 species, there were 6 species of Eulopidae indicated high parasitization levels namely *Neochrysocharis* sp., *Hemiptarsenus varicornis* Girault. *Opius* sp., *Asecodes deluchii* (Boucek), *Pnigalio* sp. and *Quaastichus* sp. The four first of them, have special characters in the field. *H. Varicornis* has high parasitization level and disperse at all locations in Bali and Lombok. Meanwhile, *Opius* sp. has high abundant population with high parasitization level at high land (especially in Baturiti, Tabanan and Pancasari, Buleleng, Bali). *A. deluchii* and *Neochrysocharis* are more specific at low land and middle altitude. The average of parasitization levels of the four parasitoids was 19.6 percent (for *Neochrysocharis*), 17.8 percent (for *H. varicornis*), 6.4 percent (for *Opius* sp.), and 6.5 percent (for *A. deluchii*) respectively. In Bali, the *Opius* sp. was mostly observed on potato crops at the high land areas, while *Neochrysocharis* sp. and *A. deluchii* were more observed on bean crops especially yard-long bean at lowland areas. *H. varicornis* was found on all host plants in mostly crop locations and altitudes in Bali and Lombok. Meanwhile, *Neochrysocharis* sp which was found in Lombok more observed on beans in lowland.

Detection of prey DNA within generalist predators: tales of beetles and spiders

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The most accurate method for determining the diets of generalist predators in the field has, until recently, involved the relatively difficult and expensive development of prey-specific monoclonal antibodies. We report here our rapid progress developing and applying prey-specific primers to amplify and detect the DNA of a range of different species of molluscs, annelid worms, aphids, Diptera, Collembola, Lepidoptera, aphids and nematodes within the guts of spiders and both carabid and coccinellid beetles. New techniques are reported that allow large numbers of field samples (predators) to be rapidly and simultaneously screened for a broad range of prey targets. Applications of this technology to the testing of important ecological hypotheses will be described, including areas such as the effects of biodiversity on biological control, prey selection, the importance of detritivores as alternative prey, classification of predators into guilds by determination factors such as when predation occurred, and infiltration of native food webs by alien species.

"Coevolution between *Wolbachia* and *Trichogramma* under local mate competition"

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Local strains infected with parthenogenesis-inducing *Wolbachia* have been found in gregarious egg parasitoid wasps, *Trichogramma kaykai* and *T. dendrolimi*. In case of *T. kaykai*, the embryonic mortality of infected individuals is much higher than that of *Wolbachia*-free ones. In contrast, *Wolbachia* infection does not significantly affect the mortality in *T. dendrolimi*. To explain the difference in the interaction between *Wolbachia* and *Trichogramma*, we developed a lattice model in which the positive effect of fertilization on the immature survival rate and the effect of female-biased sex ratios under local mate competition are incorporated. Simulation results revealed that the sex ratio has a prominent effect on the successful invasion of *Wolbachia*-infected wasps into a population. If mating occurs randomly and the sex ratio is not biased in *Trichogramma*, infected wasps can easily invade even though the immature mortality of infected wasps is high. However, if the sex ratio is extremely female-biased, then infected wasps hardly establish in a population unless the mortality cost is negligible. On these bases we discuss the coevolution between *Wolbachia* and *Trichogramma*.

"How many natural enemy species should we introduce for classical biological control?"

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I will review the history of three cases of classical and one conservative biological control of fruit tree pests in Japan to consider the scientific approach for the selection of natural enemies in biological control. The first case is biological control of the red wax scale, *Ceroplastes rubens*, a citrus pest by *Anicetus beneficus*. The second one is that of the chestnut gall wasp, *Doryocosmus kuriphilus* by *Torymus sinensis*. The third one is that of the arrowhead scale, *Unaspis yanonensis*, a citrus pest by two parasitoids, *Aphytis yanonensis* and *Coccobius fulvus*. All those pests and natural enemies were introduced from China into Japan, and the first tow cases were single species introductions and another was multiple species introduction. The last case is conservative biological control of the Japanese mealybug, *Planococcus kraunhiae*, a persimmon pest by two native parasitoids, *Anagyrus fujikona* and *Allotropa subclavata*. It was suggested that disordered introduction of all of the natural enemies availablewas was not appropriate. However, it may not be wise to persist with single species introductions. Careful study on host-parasitoid systems may be important for making introduction decisions.

"Parasitoids invasions on the leafminer *Lithocolletis Sp.* (Lepidoptera: Gracillariidae) on *Pongamia* sp. (Leguminosae) trees in Delhi, India."

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"The field study of parasitoid fauna on the leafminer host *Lithocolletis sp.* already sampled on *Pongamia sp.*, widely grown medicinal tree, has been carried out for two years (2002-04). The two phases focused during the study are:
Quantitative sampling covering random collection of a series of mines, twice a week, to determine the degree of parasitization & phenology particularly the seasonal variation of the parasitoid.
Parasitized larvae of *Lithocolletis sp.*/pupae of the parasitoids reared to eclosion to identify the species of parasitoids.
Studies reveal that the parasitoid complex includes ten species. The dominant species are *Ageniaspis fulvicornis* (Encyrtidae) and *Pholetesor sp.*(Braconidae), co-dominated by *Closterocerus insignis* (Eulophidae). *A. fulvicornis* is a larval endoparasitoid, polyembryonic, most likely as primary/secondary and a koinobiont with peak parasitization in December 2002. *Pholetesor sp.* is larval endoparasitoid, solitary, presumably primary and a koinobiont with peak parasitization in November 2002. *C. insignis* is a larval ectoparasitoid, solitary, likely as primary and a koinobiont with peak parasitization in October 2002. Thus there are seasonal variations. The percentage parasitisim of *Pholetesor sp.* and *C. insignis* declines in 2003 whereas *A. fulvicornis* is not significantly affected. Varied species richness of the parasitoid in different habitat indicates that some of the parasitoids are possibly niche specific than just host specific.
"

"Dispersion and host searching capacity in *Trichogramma*: attributes that influence their success as natural enemies"

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"The ability of adults to find hosts – host searching behaviour - plays a key role in the success of an egg parasitoid such as Trichogramma. Environmental and genetic factors will both contribute to phenotypic variation in the searching behaviour of Trichogramma. Environmental factors include the size and type of the rearing host egg, which in turn can influence parasitoid body size and parasitoid wing shape, as well as the thermal environment experienced by immatures and adults. Increased size and changes in some shape parameters are associated with increased field fitness as measured by success in locating hosts. Genetic variation in traits influencing foraging ability such as walking speed and the ability to prospect over large areas per unit time will influence searching success, but given the low heritability demonstrated for these traits, it is not clear if genetic factors make a major contribution to phenotypic variation. This in turn raises the value of screening strains for laboratory traits as a way of predicting field success.
"

An egg host unit: what is it and what does it involve.

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Parasitoids that use eggs as a host must locate and recognize the appropriate host egg. This involves a number of factors. These factors will be discussed and the factors that are important to the parasitoid make-up what will be described as a egg host unit.

Effects of modern pesticides on the predator *Chrysoperla carnea* **and the parasitoids** *Psyttalia concolor* **and** *Hyposoter didymator***. a comparison of laboratory results**

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The use of beneficial arthropods has become wide-spread in IPM programs, but pesticides still play a key role in managing many pests. Accordingly, an essential step for their joint use is to determine the impact of pesticides on the beneficial of interest. Pymetrozine (Plenum®), Natural pyrethrins+PBO (Pelitre Hort®), Triflumuron (Alsystin®), Fipronil (Regente®) and Imidacloprid (Confidor®) were tested at the maximum recommended rate in Spain (300, 80, 150, 30, 150 mg a.i./l) in residual, topical and ingestion treatments. Pupae of the enemies were not affected by any pesticide, and effects on adults were dependent on the species and the route of exposure. Pymetrozine and triflumuron were generally, the safest for the enemies, and fipronil and imidacloprid the more harmful. Pyrethrins were harmful to adults of every enemy by residual contact or topical application and much less toxic when adults fed the insecticide.

100 YEARS OF BIOLOGICAL CONTROL IN QUEENSLAND

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"Biological control programs have been carried on weeds in Queensland since the unsuccessful introduction of a cochineal beetle (*Dactylopius ceylonicus*) to control prickly pears (*Opuntia* sp.) in 1903. The subsequent successful control of a number of prickly pears in the 1920-30's set the tone for the use of this technique, both in Australia and across the globe. Not only have imported biological control agents been responsible for significantly reducing the impacts of many of Queensland's most serious weeds but the use of biological control has also entered the culture as an acceptable and desirable option for the management of weed species, if not at times overrated. Biological control programs are currently being questioned, however, after the recent off-target impacts of a biological control agent imported to control lantana (*Lantana camara*) started to attack to commonly grown garden plants and due to costs. Historical assessments of weed impacts and analyses from recent cost: benefit studies will be presented to demonstrate the significant contribution that the science of biological control has provided to Queensland.
"

Selecting agents for biological control of fruit-infesting Tephritidae

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Members of the family Tephritidae are among the most economically important pests of fruits worldwide. Several species, primarily in the genera *Ceratitis* and *Bactrocera*, have been accidentally introduced to several parts of the world, where they have become established. Biological control can be an important tool in IPM programs targeting these pests in areas where they are adventive. Selection of natural enemies for biological control involves initial discovery and acquisition of biological information essential for successful culturing of potential agents, but also must address such issues as host specificity and non-target impacts. We discuss the problems associated with selection of natural enemies of tephritids in a classical biological control setting involving foreign exploration in subsaharan Africa. The primary focus is on Mediterranean fruit fly, *Ceratitis capitata* (Wiedemann), but we also discuss other species of *Ceratitis* and *Bactrocera* found in the Afrotropical Region.

The distribution of spider families in cotton around Australia.

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"The aim of Integrated Pest Management (IPM) is to manage pest infestations in agricultural systems by using natural enemies and more selective products. The most abundant group of predators in agricultural systems are spiders, a group which has been largely overlooked in the management of pests in cotton. In this study I identified the spider communities associated with cotton in seven different locations around Australia. I established whether these communities were structurally identical (that is, whether they consist of the same guilds represented by different species); and whether the community structure changed over the course of a year. I found that spider communities were not structurally identical at different locations around Australia. In the south they were dominated throughout the year by the stalkers, Oxyopidae, while in the north they were dominated throughout the year by the foliage runners, Cycloctenidae. In more central latitudes there was a succession of different spider families dominating the community throughout the season. The dominance of different families and guilds at different sites, and the fact that some sites experience a dominance succession while others don't, have ramifications for the implementation of IPM practices at the different sites.
"

The use of the gregarious egg parasitoid *Tetrastichus coeruleus*, **a biological control agent on the aspargus beetle (***Crioceris asparagi***).**

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The asparagus beetle, *Crioceris asparagi,* causes economical damage in asparagus crops in the Netherlands. Pesticides are used to control this beetle. The parasitoid *Tetrastichus coeruleus* is its natural enemy and is able to keep beetle populations at low densities in the wild and in US asparagus crops. We are trying to set up a mass rearing of this biocontrol agent to make it available to asparagus growers. Knowledge of diapause induction and termination is essential for this goal. Last instar larvae of *T. coeruleus* diapause in a cocoon, made in the soil by the host larva. In spring, development is resumed and after pupation adults emerge. By rearing the parasitoids under different circumstances, we tested which factors are responsible for diapause induction and determined the sensible period. Light conditions during the larval development determine the percentage of larvae entering diapause. Under light conditions that resemble those of the asparagus season (16L:8D) roughly a third of the larvae enters diapause. When light conditions are 12L:12D, 75-100% enters diapause. Larvae in diapause are stored at 4°C. Diapause is terminated after 60-100 days. Diapausing larvae can be stored for at least 200 days. Storing reduces time and costs of production and facilitates distribution.

"Integration of transgenic cotton with IPM – opportunities and challenges"

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The introduction of Bt-cotton in 1996 created new opportunities and challenges for IPM in Australia. In this first commercial season, Bt-cotton crops were grown (Ingardâ Cry1Ac) with area capped at 10% of the industry. Bt-cotton reduced the need to control *Helicoverpa*, enhancing the opportunity to conserve beneficials. Bt cotton provided cotton producers a low risk starting point to experiment with IPM. The availability of Bt-cotton, increasing availability of more selective insecticides and a co-ordinated extension effort, provided the impetus for increased adoption of IPM practices by growers. Pesticide reductions of over 70% a.i. have been documented. In 2003, Bt-cotton area increased to a cap of 40% including Bollgard II (Cry1Ac & Cry2Ab), which will totally replace Ingard in 2004 with further reductions in insecticide use expected. Continued effectiveness of Bt cotton requires effective resistance management and close attention to secondary pests (aphids, mites, whitefly). The prevalence of sucking pests (mirids, aphids, jassids) has increased in transgenic crops, because these pests were formerly controlled by applications targeting *Helicoverpa*. Products for controlling mirids mostly disrupt beneficials increasing risks of secondary pests. The challenge for Australian cotton is to develop tools to selectively manage sucking pests without disrupting IPM systems in the transgenics.

Importance of short-term dispersal, or trivial movement of augmentative biological control agents

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This paper examines the importance of short-term, trivial movement of augmentative biological control agents in agricultural systems. Effective, rapid dispersal within release sites is essential for effective augmentative biological control, both in terms of determining optimal release rates and positions within crops, and in ensuring that the natural enemies that are released are able to rapidly locate and attack the target pests. Many *Trichogramma* species have been considered for augmentative control programs, and in many cases, extremely limited dispersal after release has been a limiting factor in the effectiveness of these programs. However, in other cases, such as *Trichogramma ostriniae*, dispersal within fields over relatively large distances is rapid. The implication for augmentative releases of *T. ostriniae* in corn is that low numbers of release points are required for inoculation of large areas. In other species, dispersal is slower, the wasps may be less efficient and the result is a less effective augmentative agent. We compare dispersal ability in a range of species which may be used in augmentative release programs in this paper, and show how an understanding of trivial movement is essential for planning of economically viable programs.

Effect of prey density on leaving tendency of *Orius sauteri* attacking *Thrips palmi* on an eggplant leaf

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Orius sauteri is a predator with a wide range of prey species. Since 1998 this species is in use as biological control agent of thrips. In this study its searching efficiency, its time allocation on a leaf, its leaving tendency and the attack of prey items are investigated in detail. About 78% of the encountered prey items were eaten. During 86% of the time searching for prey was concentrated on the lower leaf side where prey can be found. Patch residence times for empty leaves were different from those on leaves with thrips larvae, independent of the specific density. Walking activity was not affected by thrips density, and walking took place during 64% of the total patch time. The patch leaving tendency of *O. sauteri* after predation was analyzed by the proportional hazards model (Cox, 1972). The leaving tendency was affected by the presence of prey items irrespective of the specific density. If prey were present the leaving tendency decreased by 0.327, resulting in larger giving up times than when no prey items were present. The time since patch entry increased the leaving tendency, making sure that not much time is wasted on patches where encounters with prey items are scarce.

Bacterial microflora community associated with *Costelytra zealandica* (Coleoptera: Scarabaeidae) defined by DGGE profiles of 16S rRNA genotypes

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The bacterial microflora community associated with the digestive tract of *Costelytra zealandica* (Coleoptera: Scarabaeidae) was examined by using Denaturing Gradient Gel Electrophoresis (DGGE) of PCR-amplified 16S rDNA gene fragments and sequencing. Gel analysis indicated presence of a wide range of bacteria, with many species being non-culturable. The bacterial community in the midgut was highly variable and affected by environment and diet. The bacterial community of the hindgut was more diverse and less disturbed by external factors. The predominant bands on the DGGE gels of the hindgut samples were sequenced and identified as non-culturable, fermenting bacteria of the Clostridiales, Betaproteobacteria, Deltaproteobacteria and the CFB phylum. The most common genotypes were affiliated with novel Clostridiales. These novel Clostridiales species were present in the hindgut of all isolations in all larval instars suggesting a symbiotic relationship. The bacteria could not be located in egg samples collected from reproductive tissues indicating some form of horizontal transmission. The role of putative bacterial symbionts of *C. zealandica* will be discussed.

Screening of entomopathogenic fungi against the aphids, *Myzus persicae* and *Macrosiphum euphorbiae.*

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The screening of entomopathogenic fungi, 17 isolates of *Metarhizium* spp., 7 isolates of *Paecilomyces* spp. and 3 isolates of *Hirsutella citriformis* against the aphids had performed in Lampang Agricultural Research and Training Centre, Lampang, Thailand. The efficacy test has done by spraying suspension of 10^8 spore per ml. of fungus in completely randomized design experiments on *Myzus persicae* and *Macrosiphum euphorbiae* . Then the number of the dead aphids which have covered by fungus was recorded and calculated percent mortality by Abbott's formula. The results showed that the mortality percentage of *Myzus persicae* was from 0 to 89.47 % and *Macrosiphum euphorbiae* was from 0% to 100 %. The fungi which killed aphids from 50% up were selected and checked virulent level by calculating the LC_{50} using Factorial in completely randomized design experiments. Factorial A was fungal isolate and factorial B was 5 levels of spore concentration. The results showed that the number of the aphids killed by fungi were correlated with the spore concentration while the percent and duration of mortality depended on the fungal virulence and aphid species. The highest and lowest LC_{50} of fungi on *Myzus persicae* and *Macrosiphum euphorbiae* showed significantly different. There was 1 isolates showed very high effect on *Myzus persicae* and *Macrosiphum euphorbiae.* Its LC_{50} was 1.95×10^4 and 2.24×10^6 spore/ml respectively.

Assessment of *Trichogramma embryophagum* (Hymenoptera:Trichogrammatidae) against *Sesamia* (Lepidoptera: Noctuidae)

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The role of *Trichogramma embryphagum* (Hymenoptera:Trichogrammatidae) (reared on *Heliothis zea* Boddie eggs) as parasitoid of the sugarcane and corn stem borers *Sesamia cretica* Led. And *S.nonagrioides botonephaga* T.&B.,was assessed under laboratory and greenhouse condition .Data on seven biological parameters are presented.The laboratory results showed that *T.embroypham* reared on *H.zea* eggs parasitized 50% of *S.critica* gggs,compared to 58.5% of *S.nonagrioides* eggs.percentages of emergence (87%), female progeny (71%),deform females (2.5%), and development time (8.9d) in *T.embroypham* reared from *S. cretica* gggs did not differ significantly from those reared on *H.zea* eggs (93%,69%,3.2%,2d and 8.5d respectively).However,*T.embroypham* females reared form *S.cretica* eggs had greater body length (39mm) than females reared on *H.zea* (0.33mm). in the greenhouse conditions, *T.*embroypham parasitized an average of 56.4% of the *S.cretica* eggs masses were distributed on the corn plants).about 21.6% of the eggs were dessicated,probably due to puncturing by the parasitoid,compared with 4.1% in the control,where egg masses were distributed on the plants without release of *Trichogramma*.Results showed the potential for use *Trichgramma* to control corn stalk borers in a field IPM program.

Survey of capability of egg parasitic wasp *Trichogramma embryophagum* to control *Ostrinia nubilalis*, in Tehran area at 1999-2001

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The researchon this subject was based on inundative release at farm of Agricultural Faculty biological research in Rehran(Varamin area).Before and after releasing the Trichogramma amount of parasitoid and percentage of damage have been calculated as well as the rate of parasitism in control areas(Varamin)in different month of the years 1999-2001.Rearing of *Trichogramma* was done on host egg of *Ephestia kouhniella*. During September of 1999 the highest percentage of parasitism was about 90% while in 2001 it was 82%. In toghdar farm after 7times releasing the highest rate of parasitism had been 87.5%during August 1999.

Mass rearing of the egg parasitoids, *Trichogramma evanescens* & *T. oleae* (Hym: Trichogrammatidae)

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This study was conducted using the egg parasitoids, *Trichogramma evanescens* and *T. oleae* to evaluate its egg preference of the factitious host, *Ephestia calidella* and *E.cautella* and to investigate the influence of temperatures of 14, 22, 25, 28, 30, 33 and 35°C, 60± 5% RH and 16 hr light / day on some biological characteristies of both parasitoids populations. The results indicated that host egg preference of two *T. evanescens* races depend on the type of host egg that the parasite generations reared on it. The highest total fecundity was 117 eggs / female at 25°C for *T. oleae*. The estimated optimal temperature for fecundity was around 24°C and 19°C, for *T. evanescens* and *T. oleae*. respectively. The results showed a slight differences in adult emergence for both parasitoids species at different temperatures tested, expect at 35°C, where no emergence occurred. Results also indicted the presence of variable effects of temperatures on developmental periods of the parasitoids. The relation between temperature and development duration of both species was calculated, where the minimum and maximum developmental of *T. evanescens* were around 9 and 37°C and for *T. oleae*. were 9 and 38°C, respectively.

Mass rearing & efficiency study of *Eocanthecona Furcellata* (Wolff) (Hemiptera: Pentatomidae) for control of lepidopterus insect pests of asparagus in Thailand

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Mass rearing of predatory pentatomid, *Eocanthecona furcellata* (Wolff) (Hemiptera: Pentatomidae) was conducted at the NBCRC-Central Reginal Center, Kampaengsaen, Nakhon Pathom, Thailand. Many species of insect larvae were provived as preys of *E. furcellata* for mass rearing. *Spodoptera litura* F. (Lepidoptera: Noctuidae) was considered as the most easily available preys for mass-rearing of *E. furcellata*. Approximately 350 third-instar nymphs of *E. furcellata* were released in 0.5 acre in asparagus field at NBCRC-CRC for investigation on predatory efficiency. The result showed that the population of lepidopterus insect pests were reduced more than 69 percent in 48 hours after *E. furcellata* were released. About 500 of third-instar nymphs of *E. furcellata* were released in two locations of farmer asparagus fields at Kampaengsan district. Three weeks after released *E. furcellata* showed efficiency of control over the population of *S. litura* and *Spodoptera exigua* (HÜbner) (Lepidoptera: Noctuidae) in asparagus field.

Host stage preference and development of *Allotropa citri* Muesebeck (Hymenoptera: Platygastridae) on *Pseudococcus cryptus* Hempel (Homoptera: Pseudococcidae)

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Allotropa citri could parasitize all the developmental stages of *Pseudococcus cryptus*. In the experimental condition, however, no difference was observed in the percentage parasitism between the first and the second instar hosts, but the parasitism of the third instar and adult female hosts was lower than that of the first and second instars combined. No difference was observed in the developmental duration of this parasitoid between the first and the second instar hosts. Therefore, the first and second instar nymphs of *P. cryptus* were considered suitable host stages for *A. citri*. The developmental duration of this parasitoid at 20 and 25 °C under a short day photoperiod of 8L16D was not different from that under a long day photoperiod of 16L8D. Therefore, it is considered that *A. citri* could parasitize *P. cryptus* under a short day photoperiod as well as under a long day photoperiod if the temperature is maintained over 20 °C.

Biological control of the Mediterranean fruit fly in Israel

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Our objective: to use biological control to decrease populations of the Mediterranean Fruit Fly *Ceratitis capitata* (Medfly). The worldwide tendency is to reduce chemical control and to switch to environmentally friendly control methods. In Israel today, the control of Medfly is still based on organophosphates (mostly malathion - bait applications). Small fruit gardens and neglected groves are considered as major sources of infestation. Effective biological control may reduce the Medfly populations there therefore decreasing the infestation rate of commercial groves, requiring less chemical sprays. Furthermore, biological control is the major complement to the Sterile Insect Technique (SIT). Before the beginning of this study, three Braconid species were found on Medfly in Israel: the pupal parasitoid *Dirhinus giffardii*; and the larval - pupal parasitoids *Aganaspis daci* and *Psyttalia concolor* (complex). Since 2002, we have imported four other Braconid species: the egg – pupal parasitoids *Fopius arisanus* and *F. ceratitivorus*; the larval - pupal parasitoids *Psyttalia concolor* (complex); and *Diachasmimorpha krausii*. We have cultured each on Medfly and have begun to study some aspects of their biology. We have started to release them all over the country and are monitoring recovery by fruit sampling.

Fatty acid composition of eggs of two host species of *Trichogramma* & relationships with cold tolerance

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Fatty acid composition of insect eggs is closely related to the cold tolerance. In this work, the proportions of fatty acid in eggs of two noctuid species were compared, one active during summer, *Anticarsia gemmatalis*, and the other found during winter in Southern Brazil, *Mythimna sequax*. Eggs were obtained from laboratory rearing, stored in liquid nitrogen during 30 days and exposed to *Trichogramma pretiosum* at 21ºC. In another essay, eggs were parasitized at 21ºC and the parasitoids were stored in the pupal stage at 4ºC during 20, 30, 45 or 60 days. Parasitism on *M. sequax* and *A. gemmatalis* eggs after maintenance in liquid nitrogen were respectively 55.3% and 29.2%. The host did not influence on the parasitoid survival after pupae storage at 4ºC during 20 and 30 days, and the emergence ranged from 75.4% to 100.0%. After 45 and 60 days storage, parasitoids developing on *M. sequax* showed emergence (90.0% and 77.0%) higher than the one obtained on *A. gemmatalis* (38.3% and 13.0%). *M. sequax* eggs contain higher concentration of palmitic, stearic, oleic and linoleic acids, in relation to *A. gemmatalis*, so that the winter host contains higher nutritive resource and shows higher cold resistance of the cell membrane.

Biological control efficacy of entomopathogenic nematode, *Steinernema carpocapsae* against lepidopterous insects of Chinese cabbage under greenhouse and open field conditions in Korea

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Control efficacy of entomopathogenic nematode, *Steinernema carpocapsae* (SC) Weiser, was evaluated on the larvae of tobacco cutworm (TC), *Spodoptera litura* Fabricius, beet armyworm (BA), *Spodoptera exigua* Hubner, and cabbage webworm (CW), *Hellula undalis* Fabricius of Chinese cabbage at different spraying times under greenhouse and open field conditions in Milyang, south-eastern part of Korea. During late July to early October, control efficacy of SC against TC was in the range of 57 to 93% and 39 to 90% at greenhouse and open field, respectively. Likewise, during late August to early October control efficacy of SC against BA showed 51 to 94% and 42 to 90% between greenhouse and open field, respectively. On the other hand, control efficacy of SC against CW was in the range of 58% to 30% at greenhouse during middle September to early October. Accordingly, SC was very effective to control both TC and BA although their control efficacies were significantly depending upon air temperature while was not effective in the control of CW. Therefore, SC was considered as promising biological control agent against TC and BA during middle summer to middle fall seasons in Korea.

Potential use of *Trichogramma* as a tool in cotton IPM in Mato Grosso, Brazil.

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Considering, the spread use of *Trichogramma* in IPM programs all over the world, the objective of this study was to test the parasitism efficiency of *Trichogramma* when used to control Lepidoptera-pests of cotton and its potential to be used as a tool in cotton IPM of MT, Brazil. Two plots of one hectare each (management using *Trichogramma* and conventional management) were set up in Primavera do Leste, Campo Verde and Novo São Joaquim County, state of Mato Grosso, Brazil. From week zero (before parasitoid release) on, it was released 100,000 wasps/ha/week on the plots managed with *Trichogramma*. Both plots were evaluated every time that the parasitoids were released until harvest and the insect-pests densities, the number of non-parasitized and parasitized eggs of the cotton leafworm and of the tobacco budworm, as well as the number of parasitoids and predators were recorded. The results showed that a greater number of predators and parasitoids were found in the plots that were managed with *Trichogramma* than in the plots that were managed without *Trichogramma*. Both plots showed high rates of parasitism, achieving 100% in some points of evaluation.

Who should avoid intraguild predation: the predator or its parasitoid prey?

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Predators may feed on both herbivores and their endoparasitoids. Such intraguild predation (IGP) is often asymmetrical, imposing strong selection pressure on the parasitoid, the IG prey in the system. If so, we hypothesize that the IG prey should evolve strategies to minimize IGP. Female parasitoids may change foraging site, behavior or both in the presence of their IG predators, thus reducing their risk of predation. The study system consisted of an IG predator, *Coccinella undecimpunctata*, a parasitoid IG prey, *Aphidius colemani,* and their aphid prey/host. The parasitoid preferentially parasitized hosts on which it was reared, regardless of the presence of the predator. Confronting a predator, *A. colemani* did not leave the host plant, even when predator-free plants were available nearby. Yet lower parasitism rates occurred in the presence of predators. *C. undecimpunctata* larvae fed on parasitized aphids showed decreased prey consumption and increased handling time with increasing parasitoid age. Larvae reared on mummies rather than aphids developed more slowly and had lower pupal and adult weights. Contrary to expectations, results show that (I) the IG prey in the system does not avoid predation although it experiences significant reduction in reproductive output; and (II) the IG predator is adversely affected by feeding on IG prey.

Analysis of molecular adaptation of nucleopolyhedrovirus genes

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Genes in which nonsynonymous substitutions are fixed at a higher rate than synonymous substitutions are said to have undergone molecular adaptation, or positive selection. The primary selection pressure on a virus results from virus-host interaction. Hence, analysis of selection pressures on virus genes can potentially identify genes involved in species-specific virulence. To identify baculovirus genes that have undergone positive selection, a maximum likelihood approach was used to analyze selection pressures acting on genes of nucleopolyhedroviruses (NPVs) that have different host ranges within Lepidoptera. Alignments of 84 group I NPV genes were fitted to models of codon substitution that allow for varying selection intensity among codon sites. Evidence for positive selection was found for nine genes: *ac38*, *ac66*, *arif-1*, *lef-7*, *lef-10*, *lef-12*, *odv-e18*, *odv-e56*, and *vp80*. NPV genes that have undergone positive selection may modulate the ability of different NPVs to replicate efficiently in cells (*lef-7*, *lef-10*, *lef-12*) or to establish primary infection of the midgut (*odv-e18*, *odv-e56*) of different host species. Analysis of the baculovirus DNA helicase gene (*dnahel*) with a method that allows selection pressure parameters to vary among lineages suggests that positive selection may have occurred in *dnahel* within a clade containing BmNPV.

Differential response of a generalist egg parasitoid to alternative host species with different physical features

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Trichogramma pretiosum Riley is a generalist parasitoid that attacks many insect pests, mostly lepidopterans. However, parasitism rates vary substantially across host species. We investigated whether natal host species and age of adult wasp (<24 h and 2 days old since eclosion) affects the oviposition preference and parasitism rates of *T. pretiosum* on *Helicoverpa armigera* Hub. and *Spodoptera litura* Fab. When a choice was given, regardless of natal host and adult age, the female wasps responded differently to the eggs of the two species. However, when female wasps had no choice, they responded to both host species in much the same way. When given a choice of egg species, the productivity of *T. pretiosum* was significantly higher on *H. armigera* eggs, but they showed equal productivity when there was no such choice.

Control of spiralling whitefly *Leccanoideus floccissimus* Martin et al. (Hemiptera: Aleyrodidae) & *Aleurodicus dispersus* Russell (Hemiptera: Aleyrodidae) by entomopathogenic fungus

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The incidence of spiralling whiteflies *A. dispersus* and *L. floccissiumus* is one of biggest problems affecting ornamental crops and gardens in the Canary Islands. Several trials with different species of enthomopathogenic fungi for controlling these species of whiteflies have been performed under laboratory conditions. Results from these tests showed *Lecanicillium lecanii* and *Metarhizium anisophliae* as the most efficient species controlling *L. floccissimus*. Furthermore, a new bioinsecticide based on blastospores of the fungus *Paecilomyces fumosoroseus* (FuturEco S.L., Spain) was also evaluated. Preliminary tests with this bioinsecticide showed high efficacy against both species of spiralling whiteflies.

Mass production & utilization of the predatory stink bug, *Eocanthecona furcellata* **(Wolff) (Hemiptera: Pentatomidae)**

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The richness of the species of stink bug, Pentatomidae, is clearly proved by the existence of over 5,000 species recorded in the world. Besides , the predatory stink bug , *Eocanthecona furcellata* (Wolff) , plays an important role in ecosystem. This predatory stink bug is a predatory insect that can be commonly found on horticultural plants preying on various Lepidopterous insect pests in Taiwan. The predatory stink bug can attack many important pests such as *Pieris rapae crucivora,Spodoptera litura, Plutella xylostella* , *Spodoptera exigua et al.* in Taiwan. It will also attack other Lepidoptera , Cleoptera on vegetables , fruits , ornaments and many other crops grown outdoors , in gardens , in greenhouses and other areas. The predatory capacity and predatory response of the stink bug to various prey were measured and it was taken as an important data for releasing the natural enemy to control the pests in the field. The study was conducted for assessment of efficacy and profit for control of *Pieris rapae crucivora* and assessing the profit of control measure to release this natural enemy. The basic biology , mass production, utilization of *Eocanthecona furcellata* (Wolff) were established and would be discussed in this report.

Cultivation media for *Bacillus thuringiensis* subsp. *israelensis* Phrae strain, from waste of agro-industry

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Bacillus thuringiensis subsp. *israelensis* Phrae strain was proved to exhibit larvicidal activity against *Aedes aegypti*, vector carried dengue haemorrhagic fever. Juice in brown coconut or as called coconut milk, available in the coconut meat industry was therefore selected for the present study. Preliminary study indicated that among 8 kinds of media, cell mass from peptone glucose salt medium was 1.9709 g/l while those of plain coconut milk was 1.0128 g/l. The 18 recipes of coconut milk-based media (CM1-CM18) were then developed. It was indicated that the CM13, CM10 and CM11 recipes gave 1.7152, 1.6076 and 1.5955 g/l of cell mass, respectively. While the CM14, CM17 and CM15 recipes were the group of low cost. Considering on the amount of cell mass, cost and complication of the formula, the CM13 and CM17 recipes were subject to determine the larvicidal activity of the cell cultivated from both recipes in term of potency. The potency of candidate cells cultivated from CM13 and CM17 recipes were 908 and 10,657 ITU/mg, respectively. However, cell mass from CM13 recipe was 1.8 times higher than CM17 recipe while the cost was also106 times higher but potency of the cell from CM17 recipe was 11.7 times higher than those of CM13.

Mass-producing weed biocontrol agents through 'farming'

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The recent success of the European root weevil, *Mogulones cruciger* Hbst. (Coleoptera: Curculionidae), as a biocontrol agent for the rangeland weed, houndstongue (*Cynoglossum officinale* L.) in British Columbia, Canada, has created a large demand for the weevil among ranchers and land managers. Numbers of the weevil sufficient to meet this demand cannot be easily collected from field sites, and mass-production of the agent in the laboratory is expensive and constrained by availability of host plant material. Hence, we have initiated a project to mass-produce the agent that involves growing houndstongue as a crop in an agricultural setting, releasing the weevil into the crop, and after two developmental generations, harvesting the progeny for distribution. Experiments are currently underway to develop agronomic methods of cropping houndstongue, which include seeding time, depth and spacing, fertilizer and mulch treatments, and methods of controlling houndstongue diseases and other competing weeds. Insect responses to the different agronomic treatments also are being investigated with the goal of maximizing insect production. It is anticipated that this method of mass-production could be adopted by ranchers and land managers to produce sufficient weevils to meet local demand, and through modification, could be used to produce other biocontrol agents.

Temperature-dependent development of *Anagyrus pseudococci* on the vine mealybug

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The influence of temperature on *Anagyrus pseudococci* (Girault) development and overwintering was investigated to improve biological control of the vine mealybug, *Planococcus ficus* (Signoret), in California vineyards. We tested eight constant temperatures and show *A. pseudococci* completed development (egg to adult eclosion) between 14 and 34ºC. Development times ranged from 79.1 ± 1.0 days (14ºC) to 10.2 ± 0.3 days (34ºC). We determined optimal, maximum and minimum development temperatures as 24.7, 36.0 and 11.4ºC, respectively. The thermal constant is 231.9 degree-days. We compared these laboratory-derived temperature relationships with field activity of *A. pseudococci*. The parasitoid was reared from field-collected mealybugs between May and October. Laboratory-derived temperature-dependent development suggests there are seven to eight *A. pseudococci* generations during this period, two generations to each vine mealybug generation. Overwintering studies show that, regardless of when vine mealybugs were exposed to *A. pseudococci* (October 2001 to March 2002), emergence was concentrated over a 15 day period in early May. Results suggest that cues other than temperature are used to synchronize overwintered *A. pseudococci* adult emergence with field availability of vine mealybug. Results are discussed with respect to insectary rearing and augmentation of *A. pseudococci*.

Predation of *Alabama argillacea* (Lepidoptera: Noctuidae) by nymphs & adults of *Podisus nigrispinus* (Hemiptera: Pentatomidae) under different caterpillars densities & ages of cotton plants

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The predation rate of fifth instar nymphs and females of *Podisus nigrispinus* (Hemiptera: Pentatomidae) was evaluated at lower and higher threshold control levels for *Alabama argillacea* (Lepidoptera: Noctuidae) larvae, in three ages of cotton plants (20, 40 and 60 days). Five cotton plants were caged in field with one and three *A. argillacea* larvae per plant and one nymph or one female of this predator. Nymphs and adults of *P. nigrispinus* preyed higher number of larvae preyed with higher density of larvae and smaller leaf area in young cotton plants. Nymphs and adults this predator restrained plant defoliation bellow threshold control levels at both infestation levels and plant ages of 20 and 40 days, compared that of plants without this predator and on plants with 60 days. *P. nigrispinus* weight gain was affected when maintained in plants at more advanced development stages, in both tested densities of *A. argillacea* larvae.

Cold storage of egg parasitoids of soybean pentatomids as a method for large-scale production in biological control programs

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Hibernation is a common adaptation used by insects to overwinter in temperate and subtropical regions. Previous studies in natural conditions showed that *Trissolcus basalis* and *Telenomus podisi* hibernate as adults in Southern Brazil, so that storage in this stage can be an adequate procedure in the large-scale production of these species. In this work, adults of *T. basalis* and *T. podisi* kept during the immature stages at 25ºC were stored at 18ºC. Longevity of the hibernating parasitoids and fecundity after 180 days were evaluated. Alterations in the fatty acid composition of the parasitoids were measured. Mean longevity of stored females was 332.5 and 225.9 days, for *T. basalis* and *T. podisi* respectively, while non-hibernating females lived for 74.5 and 56.3 days. Storage of *T. basalis* did not influence its reproductive capacity; however, for *T. podisi*, fecundity decreased ca. 50% after hibernation. For *T. podisi*, content of pentadecanoic, palmitic and stearic acids decreased after storage, while the content of palmitoleic and oleic acids was higher in hibernating parasitoids. For *T. basalis*, storage did not influence the fatty acid composition, and it is concluded that this species can be stored during winter and released during the soybean season in Southern Brazil.

Storage of pentatomids eggs in liquid nitrogen for the production of parasitoids in laboratory

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The egg parasitoids *Trissolcus basalis* and *Telenomus podisi* are commonly found on soybean pentatomids. Storage of the host eggs in liquid nitrogen could be used to increase the efficiency of the rearing of these species in laboratory and their field release. This work was conducted in order to determine which host species are suitable to parasitism after storage of the eggs during six months in liquid nitrogen. Parasitism by *T. basalis* was evaluated on *Nezara viridula*, *Acrosternum pengue* and *Edessa meditabunda,* while for *T. podisi*, the parasitism was investigated on *Euschistus heros*, *Piezodorus guildinii*, *Dichelops furcatus* and *Podisus nigrispinus*. Among all species evaluated, *N. viridula*, *A. pengue*, *P. guildinii* and *D. furcatus* were suitable to parasitism after storage and the rates of emergence on these species ranged from 61.8 to 94.5%. On the other hand, *T. basalis* did not parasitize eggs of *E. meditabunda* after storage and the emergence of *T. podisi* on stored eggs of *E. heros* and *P. nigrispinus* decreased in comparison to the control. It is concluded that eggs of four pentatomid species can be stored in liquid nitrogen and used in the large-scale production of *T. basalis* and *T. podisi*.

Maximising the efficacy of a coccinellid predator in a biocontrol setting (*Cryptolaemus montrouzieri*)

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Coccinellids are utilised in biocontrol programs throughout the world, and they are commonly accepted as 'voracious' predators of mealybugs and scales. One such organism is the native Australian ladybeetle *Cryptolaemus montrouzieri*. It is frequently released against the citrus mealybug (*Plannococcus citri*) both in Australia and overseas. The success of field releases has proved erratic. We conducted field studies, over two years, comparing the success of different life stages in controlling mealybug infestations in custard apple orchards. The results suggest that the release of early instar larvae is more efficacious than releases of either adult beetles or eggs. This field data is presented in conjunction with three laboratory experiments. The first experiment investigates the importance of intraspecific predation for eggs and first instar larvae. The remaining two examine the survival and searching capacity of second instar *Cryptolaemus* when they do not hatch in direct contact with a food source. These results are used to make recommendations for the improved manipulation of *Cryptolaemus* in a field environment.

Effect of the entomopathogenic fungus, *Paecilomyces fumosoroseus* to the green peach aphid, *Myzus persicae*.

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Green peach aphid, *Myzus persicae* (GPA) infests many vegetables, including crucifer. GPA has high fecundity and grows to a large population. Usually, chemical insecticides are used to control GPA, but may cause many problems. So, it is necessary to produce other ways to control GPA, and we estimated the control effect of *Paecilomyces fumosoroseus* to GPA. We isolated the entomopathogenic fungus, *P. fumosoroseus* (strain SPf-1) from soil that indicates high virulence to GPA. In laboratory conditions, SPf-1 cause high mortality and make lots of conidia on the cadaver of GPA. SPf-1 still showed high virulence even when the concentration was reduced. Generally, entomopathogenic fungi are suitable and virulent at 20~25°C. However in Japan, GPA infest much at early spring to late autumn when the temperature is relatively low, than high temperature at summer. To determine the usefulness of entomophatogenic fungi, researches at low temperature is necessary. In this study, we estimated the efficacy of SPf-1 to low temperature conditions, and efficacy at field environment. Furthermore, to enhance the fungal virulence, and gain stability effect under greenhouse environment, bioassays using soybean oil were conducted.

Indigenous insect parasites attack the introduced pine weevil, *Pissodes nemorensis,* in South Africa

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The deodar weevil, *Pissodes nemorensis,* is an important exotic pest of pines in South Africa. It was accidentally introduced into this country from North America and was first detected in 1942. Currently, *P. nemorensis* is distributed throughout the pine growing regions of South Africa where it causes significant damage, particularly to stressed trees. Two natural enemies have been found feeding on *P. nemorensis* in the field. These include a species of *Pycnetron* (Hymenoptera: Pteromalidae) and a species of *Gobertina* (Diptera: Stratiomyidae). Neither of these parasites has previously been reported to feed on *P. nemorensis* in its native range. We, therefore, believe that they are most likely native to South Africa and that they have adapted the capacity to feed on an exotic pest species. We are currently studying the taxonomy, biology and the efficiency of parasitism of these parasites. We are also considering mass-rearing techniques on artificial media, with the aim of promoting biological control of *P. nemorensis* in South Africa.

Yeast symbionts of natural enemies: green lacewings, *Chrysoperla* spp., & the brownbanded cockroach parasitoid *Comperia merceti*

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Despite much recent interest in invertebrate-bacterial symbioses, fungal associations with insects have received relatively little attention. Even though some yeast-infected insects are economically important natural enemies, we know little about the contributions of these symbionts in natural populations, or whether symbiotic relationships are compromised during insectary-rearing, with subsequent decreases in biocontrol efficacy. Here we present research characterising symbioses between yeasts and two types of natural enemies. The yeast symbionts of green lacewings, *Metschnikowia sp.* and *Candida spp.*, were previously thought to provide missing amino acids from adult lacewings' primarily carbohydrate diet. Our data suggest that the yeasts themselves require some minimum quality diet before providing nutrients to adult lacewings. In addition, we present evidence that the yeasts in green lacewings are vertically transmitted; a novel finding that has important implications for the nature of the lacewing-yeast relationship. In a second system, we are examining the role of the vertically transmitted intercellular yeast symbionts in *Comperia merceti*, a specialist egg parasitoid of brownbanded cockroaches. These wasps have the potential to substantially suppress brownbanded cockroach populations in enclosed building settings.

Evaluation of an candidate agent, *Floracarus perrepae* (Eriophyidae) & selection of the best adapted genotype for biological control of old world climbing fern: an invasive weed in Florida, USA

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Surveys for biological control agents *Lygodium microphyllum*, an invasive fern in Florida, USA were conducted in the its native range which includes Australia, Asia and Oceania. Twenty-two herbivores were documented including an eriophyid mite, *Floracarus perrepae* Knihinicki & Boczek. Molecular tools were used to match the origin of the invasive Florida fern with populations in the native range and the population from Cape York, Queensland was found to be an exact match for two chloroplast DNA sequences. Field studies of *F. perrepae* in Queensland found that the mite was active year round peaking during periods of ample soil moisture. Predator mites and a pathogen had significant impacts on *F. perrepae* populations, but heavy plant damage was still observed. Pre-release field impact studies revealed that *F. perrepae* caused more than 50% reduction in biomass of *L. microphyllum* over a two-year period. Several genotypes of *F. perrepae* were identified from the native range using CO1 and D2 genes. Each of these populations were screened for their acceptance of the invasive Florida genotype of the fern and the Cape York genotype performed best. Host-range tests have shown that *F. perrepae* is specific to *L. microphyllum,* and therefore its release in Florida is recommended.

Establishment & dispersal of phorid flies, *Pseudacteon* spp. (Diptera: Phoridae) in Alabama for control of imported fire ants, S*olenopsis* spp. (Hymenoptera: Formicidae)

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Two species of *Pseudacteon* have been released in Alabama on the imported fire ant. *Pseudacteon tricuspis* is established in five populations of the red imported fire ant, *Solenopsis invicta,* in the southern two-thirds of the state and *P. curvatus* is established in four populations of the hybrid fire ant, *S. invicta* x *Solenopsis richteri*, in the northern third of the state. Establishment and dispersal of the flies from release sites is documented. Fire ant populations are monitored at release sites and control sites. Biological data are collected on the behavior of both species of phorid flies.

Attractiveness of flowering plants in the urban landscape to beneficial Syrphidae & Hymenoptera

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Studies have shown that the size and shape of flowers influences the types of hymenopteran natural enemies that can forage successfully in those flowers. In this study, we examined this phenomenon over a three year period to see if a similar effect could be found with beneficial insects attracted to flowers used in urban landscapes. To do so, common annual and perennial flowering plants were sampled once a week between May and September in two different locations during 2000, 2001 and 2002. In addition, flower shape and size was evaluated and correlated with attractiveness to different species. Differences in the type and abundance of the species attracted to different flower species as they relate to floral dimensions are discussed.

Biological control of tephritid fruit flies: a novel technique for efficient mass production & field delivery of the egg parasitoid, *Fopius arisanus.*

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The introduction and establishment in Hawaii of tephritid fruit flies, *Bactrocera cucurbitae, Ceratitis capitata, B. dorsalis* and *B. latifrons* causes direct damage to vegetable and fruit crops and high cost for exporting fresh produce to domestic and export customers due to quarantine treatments. To reduce the need for pesticide use, several species of natural enemies have been introduced into Hawaii to suppress tephritid fruit fly populations. The most successful of these was the egg-larval parasitoid, *F. arisanus*, parasitizing its preferred host *B. dorsalis.* Recently, we made a breakthrough in the development of a cyclindrical cage for mass producing *F. arisanus* in the laboratory and releasing it in the field. We have produced over 100,000 *F. arisanus* per week from 6 cyclindrical cages. With this tool we can mass produce (50,00-70,00) *F. arisanus* for hand carrying this parasitoid to a release site in sufficient numbers to conduct 1) rearing and colonization studies using, as host, a laboratory colony of *Bactrocera* or *Ceratitis* fruit flies; and 2) release two batches (4,000-6,000 each) of *F. arisanus* in the field at sites where fruit flies are infesting preferred hosts to rear this egg parasitoid on wild tephritid fruit flies.

Flower strip management closed to vegetable crops to enhance aphid biological control

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Introduction of herbaceous strip and set-asides closed to openfields was demonstrated to be of great interest on the entomological diversity and abundance. Whether "agro-environmental measures » are developed for several years in Belgium, flower strip seed mixture need to be improved to attract entomophagous beneficials and enhance biological control of aphids such as in vegetable crops. Plant composition of herbaceous strip was adapted by the introduction of wild plant species to attract pest predators. Four wild plant species were selected for their attractivity and the flowering period (*Chrysanthemum segetum* L., *Agrostemma githago* L., *Papaver rhoeas* L. and *Centaurea cyanus* L.). Two seed mixtures were tested in two vegetable crops, namely green pea and broad bean, and compared to conventional seed mixture of Fabaceae and Poaceae species. In 2002, more than 4000 *Syrphidae* were trapped on 14 weeks of observations. Eighteen genera and 31 species were identified, five genera constituted 89 % of the trapped individuals. Mean density of hoverflies in conventional herbaceous strip were significantly lower than the ones from both adapted herbaceous strip mixtures including the wild flowering species. In 2003, more than 2500 syrphids belonging to 15 genera and 24 species were trapped. More than 87% of them belonged to 5 genera. Yellow traps in both adapted flower strip mixtures contained significantly higher hoverfly density than in the conventional herbaceous strip mixture. Association of *C. segetum* and *A. githago* was found to be the more interesting wild species to be introduced in to attract aphidophagous hoverfly species. Flower strip closed to vegetable openfields was shown to be an efficient way to increase the aphid predator populations leading to more efficient natural biological control of insect pests such as aphids.

Functional Response of *Menochilus sexmaculata* **(Fabricius) predating on** *Macrosiphoniella sanborni* **(Gillete)**

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The functional response of *Menochilus sexmaculata* (Fabricius) predating on old *Macrosiphoniella sanborni* (Gillete) nymphs fitted the model of Holling II type well. However, the predation was affected by temperature, vessel space and prey density. In the same temperature treatment, the higher of the prey density was, the more of predation number was found and the lower of finding area was observed. In the range of 15°C - 25°C, the predation ratio was the highest at 25°C, the average number of predation was 95 capita in one day, and the highest was 392.1 capita. The relationship between predation number and temperature was fitted with quadratic regression equation. Intraspecific interference was found in the experiment. The predation number of *M. sexmaculata* decreased with the increasing of its density. The relationship between predation ration (E) and natural enemies density (P) was described by equation of $E = 0.9003P - 0.9816$. Searching constant (Q) is 0.9003, and disturbing coefficient (m) is 0.9817.

Plant & habitat composition influence foraging behaviour of the parasitoid *Diadegma semiclausum* **(Hellén)**

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The foraging behaviour of *Diadegma semiclausum* was studied in semi-field set-ups of different plant composition. The set-up consisted of a group of plants surrounding one or two target Brussels sprout plant infested by *Plutella xylostella* caterpillars. The surrounding plants were either malting barley or white mustard interspersed with sprouts, whereas the control consisted of sprout plants alone. We analysed factors influencing the arrival and leaving tendencies of the parasitoid *Diadegma semiclausum* in these set-ups. Barley and mustard decreased the arrival tendencies of parasitoids to the first target plant. The second target plant was located with the same efficiency in the different set-ups. Females in the mustard set-up tended to spend more time on target plants over successive visits. In all set-ups the total number of hosts encountered was approximately the same. However, in the mustard set-up parasitoids had to search longer to encounter hosts. Parasitoids had lower leaving tendencies from the mustard than from the other set-ups. The number of visits to target plants increased the leaving tendencies. Thus, plant species composition of habitats influence time allocation and foraging efficiency of parasitoids. Implications of these effects on host-parasitoid interactions and efficiency of biological control are discussed.

Host specific inhibition of immune reaction by entomopathogenic fungus *Nomuraea rileyi*

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The entomopathogenic fungi such as *Nomuraea rileyi* generally inhibit their host immune reaction. To convince the inhibition of host immune reaction by *N. rileyi* (Strain SdNr-1: isolated from *Spodoptera depravata*), hemocytes and its phagocytosis in the hemolymph of three spices of Noctuidae (*S. depravata, S. litura* and *Mythimna separata*) were observed after conidial injection of *N. rileyi*. The number of plasmatocytes and granular cells were decreased after injection of SdNr-1 to *S. depravata* compared with other lepidopteran larva. In the hemolymph of *S. depravata*, the phagocytosis and encapsulation by these hemocytes to FITC labeled conidia was inhibited by injection of *N. rileyi*. The phenoloxidase activity of hemolymph of *S. depravata* was reduced after injection of *N. rileyi* compared with other two spices. These results indicated the SdNr-1 had highly pathogenicity to *S. depravata* and the pathogenicity was caused by the effective inhibition of immune reaction of *S. depravata*.

Ichneumon promissorius - a pupal parasitoid of *Helicoverpa armigera*

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Ichneumon promissorius parasitises many noctuids including the pest species *Helicoverpa armigera, Helicoverpa punctigera, Mythimna convecta, Persectania ewingii* and *Spodoptera exempta*, although parasitism rates are generally low. *Ichneumon promissorius* is found predominately in south-eastern Australia and in the south west of Western Australia as well as New Zealand and islands in the south Pacific. Development takes around 18 days at 30°C, with males emerging a day before females. Fecundity of females is variable ranging from 45 to 120 eggs. Lifespan for males is 27 days; lifespan of females is variable and linked to oviposition and mated status. In an experiment with four diet treatments, male lifespan was longest (45 days) when caged on cotton plants, compared to males restricted to water only (10 days). Female lifespan displayed a similar trend with wasps living longer on cotton plants (60 days) and least with water only (27 days). Fecundity was influenced by diet; the wasps fed on honey and water had the highest fecundity (140 eggs) while wasps fed water had a lower fecundity (90 eggs). These results are discussed in relation to the impact of this pupal parasitoid on pest species abundance.

UV mutants tolerant to pesticides from strains of the entomopathogenic fungus, *Nomuraea rileyi*

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Nine strains of the entomopathogenic fungus, *Nomuraea rileyi*, were isolated from different localities in Taiwan. Two strains, designated TC and WF, were tolerant to higher temperatures and more virulent to the common cutworm, *Spodoptera litura*, and were selected to induce mutation by UV to increase virulence and tolerance to pesticides. After two-step irradiation with UV (254 nm), 55 mutants were obtained from various colonies. These mutants were assayed for virulence and tolerance to pesticides. Only six of them exhibited better virulence than the wild type against *S. litura*. Among 17 pesticides tested, only maneb, mancozeb, and copper oxychloride + mancozeb showed inhibitory effect on colony growth. Further screening of the virulent UV mutants against these three chemicals revealed that only two mutants, TC-168 and WF-245, possessed some tolerance against fungicides. However, TC-168 was not virulent to *S. litura*. Therefore, further UV irradiation of mutants and continuous screenings of strains need to be undertaken.

Complete sequence & organization of rRNA genes of *Nosema spodopterae* in Taiwan

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"The rRNA gene organization of *Nosema spodopterae* displays a similar pattern to *Nosema* type species, *N. bombycis*, that is 5'-LSUrRNA (2,497 bp)-ITS (185 bp)-SSUrRNA (1,232 bp)-IGS (277 bp)-5S rRNA (114 bp)-3'. Compared to the organization of eukaryotic rRNA genes, the *N. spodopterae* rRNA genes display a reversed position of LSUrRNA and SSUrRNA in the operon, and believed it to be an important characteristic of the *Nosema* species. The secondary structures of SSUrRNA , LSUrRNA and 5SrRNA are constructed. This investigation demonstrates several *Nosema*-specific features of secondary structure.
"

Nucleotide sequence of 3'-end of *Homalodisca Coagulata Virus-1* **new virus from the glassy-winged sharpshooter,** *Homalodisca coagulata* **(Say) (Hemiptera: Cicadellidae)**

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Nucleotide sequence was determined for the 3'-end of a new virus isolated from the glassy-winged sharpshooter, GWSS, *Homalodisca coagulata* (Say) (Hemiptera: Cicadellidae). The virus, named *Homalodisca coagulata virus* 1 (HCV-1), was isolated from adult and 5th instar nymphs. We have cloned and sequenced a 3200 bp cDNA representing the 3' -end of the HCV genome. Sequence analysis revealed that the 3' ORF encodes the putative capsid protein subunits. Deduced amino acid sequence and cleavage sites of HCV-1 showed similarities with the coat proteins of virus capsids of the insect-infecting Picorna-like viruses, within the Family *Dicistroviridae*. These are insect infecting, positive sense, single-stranded RNA viruses of 9-10 kb in size. The ssRNA Picorna-like viruses are a rapidly growing group. We propose that HCV-1 is a new member forming an independent species within the *Cripavirus* genus in the *Dicistroviridae.*

Conservation biological control of aphids in the potato field

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In the production of potato in Hokkaido, Northern Japan, aphids are the most serious pest insects due to their direct sucking damage as well as their transmission of plant viruses. Therefore, farmers usually apply insecticides 3 – 4 times on average. However, the reduction of agricultural chemicals to promote 'environmentally conscious agriculture' has been recommended in recent years. Therefore, the effect of not-applying insecticides was examined in the field for 5 years. Healthy seed tubers were used to minimize the virus diseases. The results showed that there was no outbreak of aphids observed due to suppression by indigenous natural enemies (ex. *Harmonia axyridis, Coccinella septempunctata* and *Orius* spp.); e.g. density of *Aphis gossypii*, the most harmful aphid causing direct damage, did not exceed 10 insects per compound leaf, and there were no other insect pests conspicuous. Consequently, both the tuber yield and the starch value were comparable to those in the insecticide-treated plot. These results suggest that the potato cultivation without application of insecticides has the potential to be realized.

Management of potato tuber moth *Phthorimaea operculella* (Zeller) by *Chelonus blackburni* Cameron

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The potato tuber moth, *Phthorimaea operculella* (Zeller) is the most ubiquitous pest of potato both in fields and country stores worldwide. So big is the menace of this pest that in areas where it remains unchecked, the losses in stores may go up 70 per cent Insecticidal control although being the most common method of pest control is recently been discouraged considering its adverse ecological impacts. Biological means of controlling the pest population have recently gained grounds keeping in view their long-term implication in suppressing pest population without any ill-effects on ecological balance. Amongst the various biocontrol agents used against potato, the egg-larval parasitoid, *Chelonus blackburni* Cameron has specific place to refer because of its being uniparental, parthenogenetic and having a fairly wide host range. In this perspective, the present investigation entitled "Management of potato tuber moth *Phthorimaea operculella* (Zeller) by *Chelonus blackburni* Cameron" was carried out in spring crop season (January-June) of 2001 and 2002 at Entomological Research Farm, CSKHPKV, Palampur and at farmer's field at Nagrota Bagwan. The studied revealed that PTM can be managed effectively in fields by integrating releases of *C. blackburni* @ 50,000 female adults/ha with either *Bt* @0.5% and GV @ 250 LE/ha and deltamethrin @0.005% (each treatment at weekly interval). However, under storage conditions at Palampur and Nagrota Bagwan, *C. blackburni* @ 10 adults/kg of potatoes was equally effective as GV@ 2LE/kg or *Bt* @0.5% or *Ageratum* powder @ 2cm layer.

Entomopathogenic nematodes in hydrogel formulations for pest control

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Entomopathogenic nematodes (EN) isolated from insects are considered as natural pesticides. In Poland they were investigated as agents controlling *Hoplocampa testudinea* Klug., *Delia brassicae* Bauche, *Sitona* sp. and other pests. EN are also used for protection of mushrooms in the farms from flies. The effectiveness of EN in the field depends on moisture and temperature factors. Polyacrylamide is a promising hydrogel for nematodes formulation. It improves water retention of porous soils and prevents moisture deficiency to plants. In this study applicability of polyacrylamide in preparation of *S. carpocapsae* formulation was determined. Nematodes survived in good condition in those hydrogels at 5 ^{0}C and 10 ^{0}C for 4 weeks and their pathogenicity did not decrease. Pathogenicity of *S. carpocapsae* was also investigated in pot experiments. Nematodes suspended in hydrogels were more effective against test pests at lower soil moisture than those applied in water.

Bioinsecticide assessment of plants against fall armyworm, *Spodoptera frugiperda* (Lepidoptera: Noctuidae), in Morelos state, Mexico.

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Corn, *Zea mays*, is an important food source for Latinamerican countries. Farmers control the fall armyworm (FAW) by spraying pesticides but pollution, kill of non-target species and insect resistance threats the environment and its sustainability. In 1997, we initiated a screening program aimed to find cheap, reliable and environmental-friendly natural products to control FAW. Bioinsecticide activity was evaluated at laboratory and greenhouse conditions. Plant powder was supplemented to artificial diet and larval size, weight, survival and adult emergence was recorded. More than 120 spp in > 10 families has been assessed by the program. Powder of the Caricaceae, *Carica papaya*; the Fabaceae, *Pithecellobium dulce* and *Prosopis juliflora* had bioinsecticide activity. Powder (15%, w/w) and acetonic extract (5-20%, w/v) of *C. papaya* seeds var. mamey produced 100% FAW mortality at laboratory conditions. Brake down of this acetic extract revealed that fractions 16-17 and 45 produced > 50% mortality. Aqueous extracts (5-20%, w/v) of seeds of four *C. papaya* varieties (Mamey, Maradol, Hawaian, Cera chichona) had bioinsecticide activity under greenhouse conditions. However, > 50 % corn's damage reduction was obtained with 20% aqueous extract of var. Mamey. Our results suggest that *C. papaya* seeds contain more than one molecule with bioinsecticide activity.

Scale insects & mealybugs in European greenhouses with notes on biological control

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An overview is presented of settled species and occasional intercepts of hard scale (Diaspididae), soft scale (Coccidae) and mealybugs (Pseudococcidae) in N.W. European greenhouse cultures and interiorscapes. Remarks are made on biological control practices. Approximately 40 hard scale species have been encountered in greenhouses in the area. These include common and polyphagous species as *Aspidiotus nerii, Pinnaspis strachanii and P. buxii*; rare polyphagous species, settled in specific greenhouse situations; *Aulacaspis pentagona* and *Ichnaspis longirostris* and specialist species as *Furchadiaspis zamiae* (on Cycadeae), *Diaspis bromeliae, Gymnaspis achmaeae* (on Bromeliaceae), *Diaspis echinocacti* (on Cactaceae). Sometimes highly unusual species are encountered, e.g *Selenaspidius* and *Targionia* (both tentative id.) (on Aizoaceae) as small settled populations. Also *Fiorinia coronata* was intercepted. Soft scale insects are least species rich with *Saissetia coffea* and *Coccus hesperidum* regularly encountered. Less abundant are *Saissetia oleae, Coccus viridis,* and *Protopulvinaria pyriformis.* Most common mealybugs are *Planococcus citri*, followed by *Pseudococcus viburni* and *Pseudococcus longispinus*. Also *Spilococcus cactearum, Chorizococcus brevicruris* and *Rhizoecus* spp are found. Rarer intercepts are *Chaetococcus bambusae, Ferrisia virgata, Dysmicoccus brevipes and Trionymus* sp.

Potential for biological control of *Tamarix* spp. in the United States

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Saltcedars (*Tamarix* spp.) are exotic and invasive shrubs and small trees which have degraded riparian ecosystems in the western U.S. High water consumption by extensive monotypic stands of saltcedar is of special concern among water resource managers in the arid southwestern U.S.   A leaf beetle, *Diorhabda elongata* (Chrysomelidae) is the first agent approved for release in the U.S. for biological control of *Tamarix.* Field releases of beetles from Fukang, China and Chilik, Kazakistan have defoliated 95-98% of the Tamarix within a radius of 300 to 750 m (28 to 177 ha) within 3 years after releases at 5 sites north of the 38th parallel in the western U.S. Beetles from these regions failed to overwinter south of the 38th parallel as they prematurely enter diapuase in early summer in response to short day length. Collections of *D. elongata* from Crete and Tunisia are adapted to the shorter day lengths south of the 38th parallel and overwintered in Texas and New Mexico in 2003 and 2004.

The predatory role of earwig, *Labidura riparia* **japonica (Dermaptera) to the lepidopteran pests of golf course**

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There are many turfgrass insect pests in the golf courses in Japan, especially lepidopteran pests are numerous. These pests were generally controlled using chemical insecticides. Amount of insecticides treated to golf courses resulted environmental and other problems (exp. insecticidal resistance or pollution of ground water). New control strategy such as biological control, is needed to instead of chemical insecticides. In this study, we researched occurrence and behavior of the earwig (*Labidura riparia* japonica) in the Japanese golf course and estimated the predatory role to lepidopteran pests (bluegrass webworm: *Pediasia teterrullus*, lawn cutworm: *Spodoptera depravata*). A large number of *L. riparia* inhabited in the golf course, especially around bunker and fairway areas and were actived from early spring to late autumn. We observed *L. riparia* preyed upon the adult and larvae of lepidopteran pest. *L. riparia* could move long distance during one night. In the laboratory condition, adult and larvae of *L. riparia* have high ability as predator to lepidopteran pests. From these results, *L. riparia* plays important role as a biological control agent to lepidopteran pests on the golf course.

Biological control of ambermarked birch leafminer (*Profenusa thomsoni*) in Alaska

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Ambermarked birch leafminer (*Profenusa thomsoni*) can be a significant pest of birch (*Betula* sp.) in North America but can be controlled by the parasitoid, *Lathrolestes luteolator*. *P thomsoni* was introduced into Anchorage, Alaska USA in the late 1990's and now causes significant aesthetic damage to urban birch. While not lethal, repeated severe *P. thomsoni* damage can weaken trees, allowing establishment of fatal pests or diseases. A joint project between the United States and Canadian Forest Services was initiated in 2002 to bring *L. luteolator* from Canada to suppress *P. thomsoni* populations in Alaska in a classical biological control program. In 2003, work in Alaska established the range and phenology of *P. thomsoni* in the state and work in Canada located populations of *L. luteolator* in the Northwest Territories (NWT). Parasitized larvae were collected near Ft Smith NWT, brought to Edmonton, Alberta and overwintered. In 2004 caged and open releases are planned for the Anchorage area.

In vivo study of nucleopolyhedrovirus in *Spodoptera litura*

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A virus, which posses certain features in common with nucleopolyhedrovirus, has been isolated from the diseased larvae of *Spodoptera litura*. The virus was propagated in the larvae of *S. litura* and specimens were collected after 0, 3, 6, 9, 12, 15, 18, 24, 48, 72, 96, 120 and 144-h postinoculation (p.i.) for histopathology study using electron microscope. The virus replicated in the nucleus, where the packaging of nucleocapsids and formation of polyhedra occurred, and the nuclei swelled due to overloading with polyhedrae. Abnormal feature of polyhedrae appeared among normal polyhedrae during the formation of mature virus.

Leafroller (Lepidoptera: Tortricidae) species & their parasitoids on fruit crops & shelter in Hawke's Bay, New Zealand

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Leafrollers and their parasitoids were collected from apple, grape and boysenberry crops, and shelter plants in Hawke's Bay, New Zealand. Sixteen properties were visited monthly from November or December until April or May, between 1996-99. Leafroller larvae and pupae were reared to determine their species, or if they were parasitised. The introduced *Epiphyas postvittana* comprised over 80% of leafrollers at nine sites, whereas two native species (*Ctenopseustis obliquana* and *Planotortrix octo*) dominated on six properties. The leafroller species composition changed at one site during the study, and at another property compared with samples collected 12 years earlier. In both cases, *E. postvittana* replaced native leafrollers. Leafrollers were generally most abundant in March, but numbers varied considerably amongst sites and between years at the same site. Parasitoids from seven genera in four families (Hymenoptera: Bethylidae, Braconidae, Ichneumonidae; Diptera: Tachinidae) were reared. The relative abundance of the most common parasitoid, *Dolichogenidea* spp., was positively correlated with that of *E. postvittana*. Sites where native leafrollers were more common had more of the tachinid fly *Trigonospila brevifacies*. The percentage of leafrollers emerging as adults declined from over 60% in December to about 30% in April, as parasitism increased from approximately 10% to 50% respectively. Overall, 48% of leafrollers collected from crops were parasitised compared with 14% for larvae from shelter.

Potential for improved biological control of *Helicoverpa* in Victoria

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Two species of the noctuid genus *Helicoverpa* (=*Heliothis*) are considered to be major economic pests of agriculture within Australia. *H. armigera* has an eastern coastal and northern tropical distribution, while *H. punctigera* is the more abundant and widespread species, occurring throughout the mainland. Control still relies predominantly on chemical insecticides, however insecticide resistance is becoming increasingly problematic, along with the environmental and human health concerns associated with pesticide use. Research into the control of *Helicoverpa* is now focusing on integrated management strategies. This study will address important questions of whether the biological control of *Helicoverpa* in Victoria can be improved, and whether new or established natural enemies can be successfully integrated into an overall IPM Strategy. Of particular importance to the study are the larval parasitoids *Microplitis demolitor* and *Cotesia kazak*. *M. demolitor* is an important native parasitoid, while *C. kazak* is an exotic parasitoid responsible for significant reductions in *Helicoverpa* populations in New Zealand. These species are favoured candidates for biological control in Victoria because they both parasitise early larval instars, killing the host before it completes more than 10% of its' total feeding capacity. The feasibility of using these parasitoids in biological control programs will be investigated.

Virulence & persistence of *Beauveria bassiana* for control of Fuller's rose weevil larvae & conidial persistence in kiwifruit orchard soil

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Fuller's rose weevil (FRW), *Naupactus cervinus*, has recently become a pest of kiwifruit in New Zealand because export fruit infested with FRW eggs do not meet Japanese quarantine standards. Biological control of FRW is sought as the use of chemical pesticides is restricted on kiwifruit. Eleven strains of the entomopathogenic fungus *Beauveria bassiana* and three strains of *Metarhizium anisopliae* were tested for virulence towards FRW larvae, with the greatest activity shown by strains of *B. bassiana*. The persistence of conidia in orchard soil was also assessed. Persistence was highly variable with some strains falling to background levels after one month. Other strains showed greater persistence and possible recycling, with a greater number of conidia detected after one month than the original application concentration. The implications of variation in virulence and persistence in terms of potential biocontrol of FRW are discussed.

Biological control of fruit-tree borer, ***Maroga melanostigma*** **(Wallengren) (Lepidoptera: Oecophoridae), in prune trees**

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Maroga melanostigma is an Australian native pest in many species of trees. It is of particular economic importance in prune (*Prunus domestica*) trees in which borer activity reduces productivity and can eventually kill the tree. Young, New South Wales, the second largest prune-growing district in Australia, is where trees are most seriously affected by *M. melanostigma*. Only physical control of the pest is available, which entails using wire to infiltrate borer tunnels to pierce the larva. This is time-consuming and impractical. Minimal insecticide use on prunes in this area offers good prospects for biological control. Life cycle studies are in progress in three orchards in the Young district, up to 60 km apart. Experimental techniques include: field cages filled with detached borer-infested limbs; supplemental light-trapping to detect adult emergence; determination of oviposition by caging adults on prune limbs; and challenging young trees with early instar larvae. Eggs of *Helicoverpa* sp. have been used in the prune orchards to determine background populations of predatory insects, including *Trichogramma* spp. Biological control options favour the use of *Trichogramma* spp. following successful use of *T.* nr *brassicae* (formerly nr *invelae)* against *M. melanostigma* on a pecan farm in northern NSW.

Association of insect pathogens with the crop of *Myrmeleon bore* larvae

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Fluid-feeding carnivorous insects inject their regurgitant into prey for extra-digestion. We have found that the color of insects predated by the pit-building antlion *Myrmeleon bore* changes to dark brown, a symptom resembling that of insects infected by pathogens. Since the mandibles are linked to the crop, it was postulated that insect pathogens in the crop move into prey along with the regurginant, causing such a death symptom. Thus, microorganisms were isolated from the crop of the antlion and tested for their pathogenicity in the larvae of *Spodoptera litura* by oral application as well as by injection. Some bacteria isolated killed the cutworms with darkening of the body color when injected. In addition, several *Bacillus* species as well as "injection-active" pathogens showed pathogenicity in the larvae when applied orally using artificial diets. Interestingly, a bacterial species isolated from the antlions was more pathogenic than its type cultures. All these results suggest that crop-associated pathogens are likely to utilize the prey-capturing behavior of antlions to increase the chance for their infection in compatible insects.

Host attributes influencing host range in *Encarsia formosa* Gahan

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Whiteflies (Hemiptera: Aleyrodidae) are a diverse group of insects occurring on most plant families. Adults are free living whereas the immature stages are sessile and occur on leaves. Immature whiteflies display a diverse range of cuticular architecture including colour variation, the production of various forms of wax, stacking of exuviae, presence or absence of spines, and differences in profile. The evolutionary reason behind this architecture is unknown, but these attributes may provide a barrier to predators and parasitoids. Prior studies indicate that for certain members of the parasitoid genus *Encarsia*, there is a strong correlation between polyphagy (>6 hosts) and the host cuticle being pale in colour. As not all pale-bodied nymphs are parasitised by these polyphagous species, other attributes may be influencing host acceptance. Current studies are focussing on the role of exuvial stacking and presence of flocculent wax in the whiteflies *Lipaleyrodes atriplex* and *L. euphorbiae*, respectively. Manipulation experiments will further determine whether these attributes have an effect on oviposition and foraging behaviour.

Genetic variation in the *Cotesia flavipes* complex of parasitic wasps: towards the effective biological control of stemborer pests in Australia

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The *Cotesia flavipes* species complex of parasitic wasps are economically important worldwide for the biological control of lepidopteran stemborer species associated with gramineous crops. The complex, characterised in part by a dorsoventrally flattened body, currently comprises three species: *C. flavipes* Cameron, *C. sesamiae* (Cameron) and *C. chilonis* (Matsumura), which are morphologically similar. The absence of clear diagnostic characters to separate the species and inaccurate identification have confounded past efforts to assess the impact of specific introductions. Moreover, small- and large-scale geographic populations have exhibited differences in host/habitat preference and host range. Founder effects, drift, inbreeding and adaptation to new environments can occur during the introduction and establishment of biological control agents and may promote the genetic divergence of populations. Molecular markers are being developed to characterise genetic variation and phylogenetic relationships among these and other species and among worldwide populations of *C. flavipes*, and correlate these with host and/or habitat preference. The status of *C. flavipes*-like species in Australia will be determined for the preparedness of stemborer incursion into Australia. Genetic differentiation between populations may have potentially important implications for host utilisation and thus, the diagnosis of appropriate strains for biological control against specific host species.

Life history parameters of *Palexorista zonata* (Diptera: Tachinidae): host acceptance & suitability of three lepidopteran species

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Biology of *Palexorista zonata*, one of important natural enemies of *Helicoverpa armigera* in Kenya was studied. Larvae of *H. armigera* collected from Nairobi University Kabete campus were reared in the laboratory. Emerging *P. zonata* flies were used to initiate a laboratory colony. Flies mated easily in captivity and oviposited on introduced caterpillars. Host larvae were then kept individually in containers provided with artificial diet. Their development and parasitization were observed daily. Larvae of three lepidopteran species were used as hosts: *H. armigera* along with two species of stemborers, *Sesamia calamistis* and *Chilo partellus*. The two latter species were introduced in order to assess their suitability for easy-maintenance of *P. zonata* colony. Records obtained during colony rearing as well as separate experimental sets explain some aspects of *P. zonata* biology, such as average clutch size, egg hatching time, duration of larval development, and the effect of superparasitism. Stemborer species could be used in *P. zonata* rearing, however their efficiency in terms of fly's body size and the number of puparia supported by one host were lower than that of *H. armigera*.

Oviposition preference of the tachinid fly, *Exorista japonica* (Diptera: Tachinidae): can female flies recognize suitable host instars?

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The oviposition preference of the tachinid fly, *Exorista japonica* for 5th and last instar larvae of *Mythimna separata* (Lepidoptera: Noctuidae) was investigated at different host encounter rates in the laboratory. Egg retention of host-deprived females under the conditions of low host encounter rates was also examined. *E. japonica* laying unincubated macrotype eggs directly onto host cuticles, cannot usually parasitize on soon-to moult hosts as day-1 5th instar larvae of *M. separata.* Females conditioned with either host instar larva until 24h before the experiment, were provided with a pair of those host instar larvae simultaneously. The same female was examined with a pair of the hosts for 5 times at the time interval of 1 min (shortly after the previous oviposition). The females oviposited on the conditioned instar larva at first, but tended to prefer last instar to 5th instar larva. The eggs that were retained in the uterus hatched earlier than the other eggs on hosts, and successfully parasitized on day-1 5th instar larvae. The results show that the females have oviposition preference and retained eggs increase probability for parasitization at low host encounter rates.

Seasonal occurrence of parasitoids attacking *Helicoverpa armigera* (Lepidoptera: Noctuidae) in Kenya

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Helicoverpa armigera Hubner, a serious pest of important crops in Kenya, is attacked by various natural enemies, including several species of tachinid flies. Weekly collections of *H. armigera* larvae were initiated in order to obtain information on the pest and its parasitoids occurrence throughout the year. Larvae were collected from a pigeon-pea (*Cajanus cajan*) field, maintained for the purpose, located in the Nairobi University Kabete Campus. Collected larvae were kept individually, provided with artificial diet, and their development was observed. Data on seasonal occurrence, percentage parasitism and host stage attacked were compiled for months, for the period from June 2002 to the present, and compared to data of monthly precipitation and temperature in the area. The most important mortality factor of collected *H. armigera* larvae was parasitization of tachinids, *Palexorista zonata* Curran, and *Linnaemya longirostris* Macquart. Parasitization of the both species peaked in different time of the year and total parasitism sometimes exceeded 50%. The results encouraged us for further investigations of these parasitoids as biological control agents of the pest.

Techniques for efficient mass rearing of *Serangium ryukyuense* (Coleoptera, Coccinellidae) as a biological control agent

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The ladybird beetle, *Serangium ryukyuense* native to Okinawa, southern part of Japan is considered to be a desirable biological control agent for whiteflies because it has high voracity in both the larval and adult stages, and adults are long-lived and multivoltine, producing more than 15 generations a year with no diapause under laboratory conditions. We have developed a simple, low-cost and labour-saving technique for rearing this species using artificial diets containing lyophilized drone-honeybee brood powder, autolyzed yeast and sucrose at a ratio of 5:3:2. This diet successfully sustained more than 20 generations and addition of methyl-*p*-hydrobenzoate at 1% prevented moulting during the larval period. The rearing cages were simple and cheap. Cannibalism was reduced by corrugated cardboard. A method of easy egg collection using yarn as an oviposition substrate was developed. The possibility of adult storage was also studied; adults survived more than 5 months at 7° or 15°C at 12L/12D and resumed normal oviposition when the temperature was increased to 25°C, demonstrating the potential for long-term, low-temperature storage of this species.

Life history of *Euselasia bettina* and *E. chrysippe* (Lepidoptera: Lycaenidae: Riodininae) on *Miconia calvescens* (Melastomataceae) from Costa Rica

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The life history and early stages of *Euselasia bettina* (Hewitson, 1869) and *E. chrysippe* (Bates, 1866) on *Miconia calcvescens* Schrank & Mart. ex DC., are described and illustrated. The species possess six larval instars and the larvae are processionary throughout their development. The eggs are laid in clusters on the underside of leaves—the mean number of eggs in each cluster: *E. bettina* 89 (SD=9; range=82-99; n=3) and *E. chrysippe* 70 (SD=20; range=44-113; n=29). The larvae are gregarious foliage consumers that feed, rest, and molt synchronously. A speices of Aphelinidae was reared from eggs of these two species, and a species of *Calolydella* sp. tachinid fly was reared from late instar larvae of *E. chrysippe*. *M. calvescens* is currently registered as one of the threatening invasive weeds in Tahiti and in Hawaii, and these two *Euselasia* species are currently considered as one of the potential biological control agents (Miconia Project Hawaii-University of Costa Rica 2004). This information will be useful for the biological control workers and personnel responsible identifying insects in quarantine in Hawaii.

Purification & functional characterization of insecticidal sphingomyelinase C produced by *Bacillus cereus*

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A group of antlions, the larvae of lacewing Myrmeleontidae, makes cone-shape pits to capture prey. When antlions bite prey, they inject regurgitant including acute toxic factors into the prey to anesthetize it. We have found that *Bacillus cereus* isolated from the larvae of *Myrmeleon bore* secretes proteins that paralyze and kill insects when injected. One of these active proteins was purified from the culture broth of *B. cereus* using anion-exchange and gel-filtration chromatographies to a single polypeptide with a molecular mass of 34 kDa. The purified insecticidal protein was able to paralyze German cockroaches, *Blattela germanica*, within a short period after injection. On the basis of the N-terminal and internal amino acid sequences of the purified protein, the toxin was identified as sphingomyelinase C. A recombinant sphingomyelinase C expressed in *Escherichia coli* was as potent as the native protein in killing the cockroaches. Site-directed mutagenesis (His151Ala) that reduced the maximum catalytic rate of sphingomyelinase C also abolished the insecticidal activity, suggesting that the rapid insect toxicity of sphingomyelinase C results from its phospholipid-degrading activity.

Differences in distribution of *Orius* spp. in relation to flowering height of plants

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Anthocorid bugs *Orius sauteri*, *O. strigicollis*, and *O. minutus* are common species distributed in mainland Japan. Although these bugs can be found on a variety of plants, especially on these flowers, the microhabitat for each species seems to be different. While *O. sauteri* adults were abundant on flowers at lower than 20cm above the ground such as white clover, *O. minutus* were mainly found on flowers at higher than 200cm above the ground such as chestnut and apple trees. *O. strigicollis* has a narrow distribution area compared to the other 2 species. In the regions where *O. strigicollis* is distributed, this species tended to inhabit the flowers at intermediate height above the ground between 20cm and 200cm. This preference seems to have no relation to families to which plants belong and pray species for *Orius* spp. inhabited on the plants. The habitat segregation observed in this study might make the coexistence of *Orius* spp. possible without severe competition.

Identification of tachinid parasitoids (Diptera: Tachinidae) of Nearctic *Choristoneura* species using puparia (Lepidoptera: Tortricidae)

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Leafrollers of the genus *Choristoneura* (Lepidoptera: Tortricidae) comprise 17 species in the Nearctic region and include such serious agricultural and forestry pests as the spruce budworm (*C. fumiferana*), western spruce budworm (*C. occidentalis*), jack pine budworm (*C. pinus*), obliquebanded leafroller (*C. rosaceana*) and large aspen tortrix (*C. conflictana*). Twenty-seven species of Tachinidae have been recorded as parasitoids of Nearctic *Choristoneura* species, of which 15 are commonly or occasionally reared and 12 represent rare or accidental parasitism or erroneous records. A paper is under preparation to provide identification keys to the adults and puparia of the 15 tachinid species in the former group, and will include a review of the biology of each species. These tachinids belong to six tribes in the Exoristinae and Tachininae and display a wide range of parasitic attack strategies, though all parasitize late instar *Choristoneura* larvae. Tachinid puparia obtained from parasitized *Choristoneura* have distinctive differences in the posterior spiracular discs that permit the species to be identified, thereby obviating the need to rear the tachinids to the adult stage. A key to, and illustrations of, the puparia of the 15 common or occasional tachinid parasitoids of Nearctic *Choristoneura* species are presented.

Identification & distribution of proteinases & other enzymes along the midgut of *Bracon hebetor* (Hymenoptera: Braconidae)

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Bracon hebetor is a gregarious ectoparasitoid and the understanding of *B. hebetor*'s digestive process and the identification and evaluation of the relative importance of its midgut proteinases could be used for development of artificial diets for this insect. The results suggest a homogeneous pattern of enzymes activity distribution. To estimate the relative contribution of proteinases to the total activity, midgut samples were applied onto a High Q column (Econosystem-Bio Rad) equilibrated with 20 mM Tris HCl buffer pH 7.5 and eluted with a linear gradient from 0 to 0.6 M NaCl and 1 M NaCl in the same buffer. The eluted fractions were assayed with fluorescent FITC-casein in the presence and absence of activators and inhibitors and also with synthetic substrates designed for each proteinase class. The following compounds were used as activators or inhibitors: 17mM SBTi, 500mM benzamidine, 100mM chymostatin, 4mM EDTA, 2mM DTT, 1mM TLCK and 40 mM E-64. The synthetic substrates used were ZFR-7-amido-4-methylcoumarin (trypsin), Suc-AAF-7-amido-4-methylcoumarin (chymotrypsin) and e-amino-caproyl-S-benzyl-cysteinyl-7-amido-4-methylcoumarin (cysteine-proteinase). The results showed that trypsin is the most important digestive proteinase in *B. hebetor*. It is responsible for almost all the protease activity upon FITC-casein. No cysteine-proteinase activity was found and the chymotrypsin activity detected was negligible.

California's avocado thrips, *Scirtothrips perseae* Nakahara: why is it a non-pest in its native home of Mexico and Guatemala ?

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The avocado thrips is just one of several recent invasive species that have challenged California's existing IPM programs. *S. perseae* was new to science, only receiving its scientific name in 1997, shortly after its arrival in southern California. Its origin was suspected to be Mexico or Central America. Subsequent foreign exploration by this author and Dr. Mark Hoddle at UCR confirmed it native range to be Mexico and Guatemala. A severe pest of southern California avocados, *S. perseae* was never recorded as a significant pest of avocado in its native home. Research conducted in these countries confirmed the presence of breeding populations of avocado thrips within commercial Hass orchards. However, these populations never reached the astronomical levels experienced over the last 5 years in southern California. Additionally, the Mexican and Guatemala populations were confined to the new growth, never moving to the tender new fruit to cause feeding scars as it does in southern California. Although there were some differences between the thrips populations in Mexico and in Guatemala, they remained confined to the flush growth, their preferred feeding substrate, which continued through the fruit set and early development periods. In the native subtropical home of the avocado, fruit set occurs during the growth flush period, while in the cooler, temperate climate of southern California avocado fruit set is delayed by 2 months. This results in the major growth flush period ending just as avocado thrips populations have peaked and young avocado fruitlets are developing. Under these conditions in southern California the thrips move from the hardening and maturing foliage to the succulent fruitlets where they continue to feed and breed, causing significant fruit rind scarring.

The role of egg parasitoides in the IPM of sunn pest in Romania

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Sunn pest (*Eurygaster integriceps)* presents a special importance for winter wheat crop. At present, the crop protection is achieved by the chemical method: about seven million ha. all over the world, from which approximately 600,000 ha. in Romania. The biological control implementation are not finished yet. Currently, the oophagous parasites develop numerous populations (30-55% parasitisation levels), but cannot ensure the sunn pest populations control. The following oophagous species are identified: *Telenomus chloropus, Trissolcus grandis, T. simoni, T. rufiventris, T. pseudoturesis, T. semistriatus, Anastatus bifasciatus, Ooencyrtus telenomicida,* the first two species being important and largely extended. The control treatments strongly distroyed the oophagous populations too. The research emphasized the best protection ways of oophagous natural fauna and their integration in the sunn pest management. During the sunn pest egg laying, the oophagous develop only 1.5-2 generations. The parasitisation level is ensured by the hibernating oophagous generation. By the diminution of the treated areas against sunn pest adults, the efficiency of parasitisation natural background is ensured. The period of oophagous embryonary development is greater than that of sunn pests. The correct application of treatments against sunn pest nymphae protects the parasites unreared yet. They constitute the natural reserve for the next year.

DAFF's procedures for biological control agent applications

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The Australian Government Department of Agriculture, Fisheries and Forestry (DAFF) is responsible for approval of the importation under the *Quarantine Act 1908*, for host-specificity test list and for release of biological control agents for the control of weeds and invertebrates. When assessing biocontrol application, Biosecurity Australia (DAFF-BA) consults widely with stakeholders including commonwealth and state/territory government departments and research organisations. BA provides recommendations to the Australian Quarantine and Inspection Service (DAFF-AQIS). If the recommendation is accepted, AQIS issues a permit for the importation of the agent into a quarantine approved premise and sends a letter of approval for host specificity test list and for release of the agent from quarantine. The Australian Government Department of the Environment and Heritage (DEH) also regulates the import of live species under the *Environment Protection and Biodiversity Conservation (EPBC) Act 1999*. If a DEH permit is required, the AQIS' permit can not be acted upon unless accompanied by a DEH permit. DAFF's procedures for dealing with biological control agent applications have been developed in consultation with DEH. The detailed process is available at www.daff.gov.au/plantbiosecurity, which include descriptions of information requirement and processing of the applications, a flowchart showing the procedure and an electronic application from. A joint application form is used by DAFF and DEH

Egg load of lab-cultured *Anaphes iole* Girault (Hymenoptera: Mymaridae)

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Anaphes iole Girault (Hymenoptera: Mymaridae) is a solitary egg parasitoid of *Lygus* bugs (Heteroptera: Miridae) in North America. Ongoing research is considering factors that might enhance the in-vivo rearing of *A. iole* using *Lygus hesperus* Knight as host. The following hypotheses were tested: (1) egg load was related to body size and not affected by female age, and (2) egg load depletion was not affected by mate presence and time (in days) that females were exposed to hosts. Initial egg load averaged 48 mature eggs and egg load was neither related to body size (hind tibia or forewing length) nor affected significantly by age (0, 1 or 2 d old honey-fed females). Mate presence (females with or without males) and exposure time (1, 3 or 5 d on the same host patch) had no effect on egg load depletion. The results of this study suggest that *A. iole* females are certainly pro-ovigenic and initial egg load does not correlate with body size or age. Since mated and unmated females deplete most of their egg load in 24 h, time-efficient production of progeny may result when ovipositing parasitoids are exposed to suitable hosts for just a few days.

Rodolia cardinalis Mulsant in Galapagos: evaluation of the first biological control program in Galapagos

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The cottony cushion scale, *Icerya purchasi* Maskell, was first reported in Galapagos in 1982. Within a few years, this cosmopolitan pest spread to more than 15 islands threatening more than 80 native plant species as well as indirectly threatening several endemic insect species by critically endangering their host plants. After a thorough risk assessment study that included feeding range studies, *Rodolia cardinalis* was released in 2002 as the first biological control agent intentionally imported to control an alien invasive species in Galapagos. More than 1800 coccinellid beetles were released on 15 islands. A long-term monitoring system was developed to determine if *Rodolia cardinalis* is able to establish in the various Galapagos habitats and if it exclusively feeds on the cottony cushion scale. Two years after the first release, *Rodolia cardinalis* has established at most release sites and has actively spread to at least one island where no release was conducted. Evaluation of predation on *Icerya purchasi* at specific sites shows a significant reduction of scale populations and subsequent recovery of plant populations. However, the likely permanent presence of the cottony cushion scales in Galapagos requires a long-term control and monitoring program including a continuous rearing of *Rodolia cardinalis*.

Impact of transgene products in honeydew & honeydew quality on parasitoid survival

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When phloem-feeding insects feed on a genetically modified plant, the honeydew can contain amounts of the transgene product as has for example been reported for plants expressing snowdrop lectin (GNA). This could pose a risk for honeydew-feeding insects such as parasitoids. Our studies show that GNA dissolved in a sucrose solution has a direct toxic effect on adults of the aphid parasitoid *Aphidius ervi* (Hymenoptera: Braconidae). However, in the case of GNA-contaminated honeydew the negative impact of the toxin is likely affected by the nutritional quality of the honeydew, as honeydew in itself is often a poor food source relative to a sucrose solution. We have investigated the suitability of honeydew from two aphid species (*Sitobion avenae* and *Rhopalosiphum padi*) feeding on GNA-expressing or control wheat plants as a food source for *A. ervi*. In addition, the honeydew was analysed for its GNA content, and carbohydrate and amino-acid composition; factors that are of importance for the nutrition of honeydew-feeding insects.

Comparison on efficiency of *Trichogramma embryophagum* & *T. brassicae* egg parasitoids of carob moth

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The carob moth, *Spectrobates ceratoniae* is a seriuos pest which has a specific feeding habit on pomegranate fruit between the green husk and the brown in all orchards in Iran. Because conventional control methods have proven ineffective, current emphasis is on the introduction and release of host-specific biological control agents. Rate of parasitism by two wasps *Trichogramma embryophagum* and *T. brassicae* was studied on densities of 5, 10, 20, 40 and 80 carob moth eggs at two different places: fruit crowns and on the same size white papers at 25±3ºC, 14 hours photoperiod and 65±5%RH in Guilan university. The results indicated that the number of hosts parasitised by *T. embryophagum* was higher than *T. brassicae* in all conditions. The percentage of eggs parasitised by both parasitoids on the papers was considerably higher than on the fruit crowns in all densities. The mean percentage parasitism of the carob moth eggs caused by *T. embryophagum* and *T. brassicae* reached to 74±2.9 and 47±3.1% respectively at density 10 on paper. Meanwhile, it was 47.5±2.7 and 24.5±3.5% respectively for two species at density 20 on fruit crown. Our results indicated that two parasitoid species could access eggs of *S. ceratoniae* in pomegranate fruit crown. However, *T. embryophagum* seems to be the most efficient species to control the pest than *T. brassicae* in the field.

Fungus derived from hindgut of passalid beetle

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A species of *Leidyomyces* was discovered from *Aceraius helferi* (Coleoptera: Passalidae) collected in Chiang Mai, Thailand. A pair of the beetle was collected from a decayed log. Hindguts of the beetles were derived by dissection. Linings of the hindguts were observed after rinsing with distilled water. Several tufts of no-branching hyphae that were sharing a holdfast (attaching structure) were observed. This fungus was observed only at the former part of hindguts. The tufts were consisted of over fifteen hyphae growing up to 750um. This fungus was identified as a genus *Leidyomyces* by both its sharing feature of holdfast and living site of the gut. Diameter and length of the holdfast was 11-25um and 11-32 um, respectively. Distal part of thallus was septate and showed a series of cell: length x diameter= 26-41 x 14.5-18 (um) average 33.5 x 15.8 (um), (N=16). Each of the cells produced a spore (primary infestation sporangiospore) inside; the genus *Leidyomyces* is consisted of only one species *L. attenuatus*. The size of the spore of our specimens was larger comparing with that of *L. attenuatus*.

Selection of insect & fungal agents for the biological control of *Emex* species within Australia

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The annual weed species, *Emex australis* and *Emex spinosa* (Polygonaceae) are approved targets for biological control in Australia. *Emex australis* originates from southern Africa and *E. spinosa* from the southern Mediterranean region. The insects associated with these plants have been surveyed in their regions of origin as well as within Australia and a range of insects and fungi assessed as potential biological control agents. Three insect species, *Apion miniatum*, *Lixus cribricollis* and *Perapion antiquum* have been tested for host specificity and subsequently approved and released in Australia. Despite concerted efforts and some massive releases, all have failed to establish within grain growing regions, the area where the weed is a problem. The reasons postulated for this have included predation by ants, suboptimal host plants, summer temperatures being too high, the absence of suitable summer refuge sites and the lack of alternative food sources in summer. This poster summarises the likely causes for lack of establishment of each potential agent and develops criteria for selection of any future agents being considered for these weeds.

The biology of *Psyllaephagus bliteus* (Hymenoptera: Encyrtidae), a parasitic wasp that attacks the red-gum lerp psyllid (Hemiptera: Spondyliaspididae)

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Psyllaephagus bliteus Riek (Hymenoptera: Encyrtidae) was released throughout California in 2000 and 2001 to control the red-gum lerp psyllid, *Glycaspis brimblecombei* Moore (Hemiptera: Spondyliaspididae). A *Eucalyptus* feeder native to Australia, *G. brimblecombei* was first discovered near Los Angeles in 1998 and had spread throughout the state by 2000. The experiments discussed herein were designed to collect basic biological information on *P. bliteus* that would supplement and improve the control program. Our specific objectives were to determine adult longevity and fecundity, to determine the larval development strategy, and to determine host-stage preference for oviposition and host-feeding. We found that although adults can live several months in the glasshouse, maximum fecundity occurs shortly after emergence. Females live longer than males at all temperatures studied. Host-feeding is common and inflicted upon nymphs of all stages. Eggs are laid into nymphs of any age but larval development begins after the host reaches the 5th instar. Most eggs are recovered from 3rd instars. However, oviposition is attempted equally into 3rd, 4th, and 5th instars. Observations of host-handling behavior suggest that the large lerps of 4th and 5th instars impede oviposition and thus protect older nymphs from parasitism by *P. bliteus*.

Utilization of *Eocanthecona furcellata* (Wolff) (Hemiptera:Pentatomidae) for augmentative biological control of the lemon butterfly, *Papilio demoleus* (Lepidoptera: Papilionidae) in Thailand

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Utilization of *Eocanthecona furcellata* (Wolff) for augmentative biological control of the lemon butterfly *Papilio demoleus* was investigated in citrus orchard.Two plots of citrus orchard were selected as control and released plot. About 1,000 third-instar nymphs of *E .furcellata* produced from NBCRC-CRC, Kampaengsan, Nakhon Pathom were released every month in released plot covering an area of 8 acre at Suphanburi province. After inoculative release the population of *E .furcellata* developed well and increased to higher levels than in control plot, while the population of *P .demoleus* decreased to lower level than in control plot. Two months after released *E. furcellata* showed satisfactory evidence of control over the population of *P. demoleus* in the released plot.

Diversity of parasitoid fauna of *Liriomyza* spp. (Diptera: Agromyzidae) on vegetable crops in Bali & Lombok

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Detailed surveys for parasitoids of the polyphagous agromyzid leafminers, *Liriomyza huidobrensis* and *L. sativae*, on a range of vegetable crops in Bali and Lombok were conducted from June 2001 until October 2003. The most abundant of the 14 parasitoid species recorded from sampled leaves were *Hemiptarsenus varicornis* Girault, *Opius* sp., *Neochrysocharis* sp. and *Asecodes deluchii* (Boucek). *H. varicornis* parasitised both *Liriomyza* spp. and was recorded from a wide range of host plants in most cropping locations and altitudes (10-1200 m a.s.l) in Bali and Lombok. *Opius* sp. was mainly observed on *L. huidobrensis* infesting potato crops in highland areas (>1000 m) (especially in Baturiti, Tabanan and Pancasari and Buleleng), while *Neochrysocharis* sp. and *A. deluchii* were more frequently observed on bean crops infested with *L. sativae*, especially yard-long bean, at lowland locations.

Coevolution between *Wolbachia* & *Trichogramma* under local mate competition

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Local strains infected with parthenogenesis-inducing *Wolbachia* have been found in gregarious egg parasitoid wasps, *Trichogramma kaykai* and *T. dendrolimi*. In case of *T. kaykai*, the embryonic mortality of infected individuals is much higher than that of *Wolbachia*-free ones. In contrast, *Wolbachia* infection does not significantly affect the mortality in *T. dendrolimi*. To explain the difference in the interaction between *Wolbachia* and *Trichogramma*, we developed a lattice model in which the positive effect of fertilization on the immature survival rate and the effect of female-biased sex ratios under local mate competition are incorporated. Simulation results revealed that the sex ratio has a prominent effect on the successful invasion of *Wolbachia*-infected wasps into a population. If mating occurs randomly and the sex ratio is not biased in *Trichogramma*, infected wasps can easily invade even though the immature mortality of infected wasps is high. However, if the sex ratio is extremely female-biased, then infected wasps hardly establish in a population unless the mortality cost is negligible. On these bases we discuss the coevolution between *Wolbachia* and *Trichogramma*.

Development & reproductive ability of *Lysiphlebus japonicus* parasitizing the apple aphid *Aphis spiraecola*

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Host age specificity, developmental periods, age-specific survival rate and fecundity of *Lysiphlebus japonicus* Ashmead were determined in the laboratory to evaluate its potential abilities as a biological control agent against the apple aphid *Aphis spiraecola* Patch. An apple leaf with ten aphids of each instar was given to a female parasitoid. Aphids stabbed once by the parasitoid were transferred on an apple seedling in a test tube (40mm in diameter, 120mm in height), which was kept in a constant temperature condition until the mummification. Mummies were enclosed individually in a small glass vial, and the date of adult emergence was recorded. Newly emerged female was mated and given sufficient number of 1st instar aphids every day for the life. All the hosts were kept rearing, and the number of mummified individuals were counted for the estimation of daily number of oviposition. As the results, *L. japonicus* attacked all stages of *A. spiraecola* and no difference was detected in the developmental period and survival rate of offspring among host ages at oviposition. Adult female survived average 4.2 days at 20'C and total 220 eggs were laid during the period. Age-specific fecundity was highest at the first day of the adult life.

Geographic variation in the abundance of *Aphidius ervi* & *Praon pequodorum* (Hymenoptera: Braconidae), parasitoids of the pea aphid

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Aphidius ervi (Hymenoptera: Braconidae) was introduced to the United States in the late 1950's and early 1960's for biological control of pea aphid [*Acyrthosiphon pisum* (Hemiptera: Aphididae)], and is now widely distributed in North American alfalfa, often achieving high levels of parasitism. *Praon pequodorum* (Hymenoptera: Braconidae) is a native aphid parasitoid which also includes the pea aphid in its host range. Prior to the introduction of *A. ervi*, *P. pequodorum* was a common parasitoid of pea aphid in alfalfa in the southern part of the State of Wisconsin. Since the establishment of *A. ervi* in this region, *P. pequodorum* abundance in alfalfa has decreased dramatically, though we lack information from other habitats and other parts of the state. Here, we examine geographic and habitat variation in the relative abundance of these two parasitoids, and discuss implications for competition and indirect non-target interactions between them.

The egg parasitoids of the brown marmorated stink bug, *Halyomorpha halys*, & the factors influencing its complex structure & dynamics

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The egg parasitoids of the brown marmorated stink bug, *Halyomorpha halys*, was examined. In parasitoids collected from egg masses of *H. halys* set on various regions, places and seasons, the *Trissolcus* species (Hymenoptera: Scelionidae) were overwhelmingly dominant, though their ratios varied. Most of these species collected from *H. halys* were also collected from *Plautia crossota stali*, whose habitat is similar to *H. halys*. *T. platuiae* in particular, was a dominant parasitoid of both the stink bugs. In parasitism on *H. halys* egg masses, this wasp exhibited a variation. *T. platuiae* strains collected from *H. halys* egg masses showed a higher performance on *H. halys* egg masses than those from *P. c. stali* egg masses. In addition, it was found that wasps reared on *H. halys* eggs increased the success rate of parasitism to *H. halys* eggs compared with those reared on *P. c. stali* eggs. On the other hand, the wasp always showed a high adaptability to *P. c. stali* regardless of its origin or development source. This result suggests that the parasitism of *H. halys* eggs is affected by the population dynamics of *P. c. stali*, which has a higher reproductive rate.

Evaluation of entomopathogenic nematodes for control of *Corythucha ciliata* Say (Rhynchota, Tingidae) overwintering adults – preliminary note

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Corythucha ciliata Say (Rhynchota, Tingidae) is a dangerous pest of plane-trees (*Platanus occidentalis* Listen). In Italy plane-trees are often located in urban areas where the chemical control can cause problems not only for the environment but also for humans. For this reason the potential of 3 Italian entomopathogenic nematodes (EPNs), as biological control agents for *C. ciliata,* was evaluated through both laboratory and field experiments. In the laboratory *Steinernema feltiae* (ItS-CL2), *S. carpocapsae* (ItS-MR7) and *Heterorhabditis bacteriophora* (ItH-CE1) were compared in petri dishes lined with filter paper against overwintering adults of *C. ciliata*. All nematodes produced high mortality rates. *S. carpocapsae* and *H. bacteriophora* produced significantly greater adult mortality rates than did *S. feltiae*. Efficiency of the EPNs was then evaluated in field trials, spraying EPN suspensions on tree trunks, where the adults of *C. ciliata* usually spend the winter under the bark. Among all the nematodes the mortality percentages of *C. ciliata* in field conditions were quite low when compared with those in the laboratory. *H. bacteriophora* produced a significantly greater adult mortality rates than did *S. carpocapsae* and *S. feltiae*. Although more studies and experiments are needed to improve the EPN potential in field conditions, these preliminary results showed that EPNs, and *H. bacteriophora* in particular, are promising candidates for further evaluation as biological control agents against *C.ciliata* overwintering adults.

Population dynamics of the diamondback moth, *Plutella xylostella* (Lepidoptera: Yponomeutidae) & it's parasitoids in Kyushu, southern part of Japan

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Population dynamics of the diamondback moth (DBM) and its parasitoids were studied for 4 years in two cabbage fields. One of the fields was set up in the net house (12 meters square and 3 meters high, screened on four sides and top by 9mm-meshed net) which prevented crucifer pests such as white butterflies and common cutworm from injuring the cabbage. Another field was maintained with standard farming practice. Cabbage plants were grown 5 to 6 times per year at each field. Population dynamics of DBM and its parasitoids were monitored twice a month in warm seasons and once in winter. Two populations of DBM showed same pattern of seasonal occurrence. DBM densities rapidly increased in March or April, and reached the highest level in May. Thereafter, DBM populations declined to the brink of extinction in June. Two species of parasitoids, *Diadromus subtilicornus* and *Cotesia plutellae* were found as dominant natural enemies of DBM. Percent parasitism by *C. plutellae* and *D. subtilicornus* in May of 2000-2003 were 36-61% and 65-87%, respectively.

Experimental release of a parasitoid, *Dastarcus helophoroides* (Fairmaire) (Coleoptera: Bothrideridae), on *Monochamus alternatus* Hope (Coleoptera: Cerambycidae)

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I conducted two release experiments of *Dastarcus helophoroides* for the biological control of *Monochamus alternatus* that is the primary vector of the pinewood nematode, *Bursaphelenchus xylophilus*. First, parasitoid eggs were released on logs that were infested by *M. alternatus* in outdoor cages as a preliminary experiment. Percent parasitism totaled 49.7 %, which greatly differed among the logs. Survival of *M. alternatus* was 45.9 % in logs onto which *D. helophoroides* eggs were released, while that in control logs was 96.4 %, which confirmed the effect that *D. helophoroides* can have on *M. alternatus* survival. When many *M. alternatus* adults had already emerged before the hatching of the released eggs began in the logs, the percent parasitism tended to be low. Second, *D. helophoroides* adults were released on dead pine trees in a stand damaged by pine wilt disease. Mean percent parasitism in 2002 and 2003 was 43.3 %. Survival of *M. alternatus* was 21.6 % in the released trees and 85.0 % in control trees. Percentage of dead *M. alternatus* by unknown factors reached 35.1 %, which was estimated to include many individuals that were parasitized but the parasitoids were dead in early developmental stage.

Quantitative evaluation of the predatory bug, *Orius strigicollis* (Heteroptera: Anthocoridae) as biological control agent against thrips

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A simple analytical model was constructed to evaluate the effectiveness of released natural enemies on annual crops. This model was applied to the life history parameters of the predatory bug, *Orius strigicollis*. The model used the ratio of insect pest and natural enemy, just when the pest population began to decrease. The result showed that the predator, *O. strigicollis*, suppressed more thrips population in low (17ºC) and high (29ºC) temperatures than in intermediate (20-26ºC). In a grower's greenhouse where green pepper was grown, the population of thrips and the released *O. strigicollis* were investigated. Three species of thrips, *Frankliniella intonsa*, *Frankliniella occidentalis*, and *Thrips palmi*, were found. When the pest population began to decrease, the pest-natural enemy ratios were compared with that of the pre-evaluation ratios. The result showed that *O. strigicollis* population at the ratio of 70:1 (active stage of thrips/adult bug) ensured the decrease in the population of thrips. Depending on complex situations in growers' greenhouses, the quantitative evaluation and usage of *O. strigicollis* will be discussed.

Effects of host diet on the fitness of *Microctonus hyperodae* Loan (Hymenoptera: Braconidae), a parasitoid of the Argentine stem weevil, *Listronotus bonariensis* (Kuschel) (Coleoptera: Curculionidae)

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The argentine stem weevil (*Listronotus bonariensis* (Kuschel) (Coleoptera: Curculionidae)) is a South American species which is New Zealand's most damaging pest because of ryegrass damage. It has been estimated to cost the pastoral industry between NZ$78-251 m per year. The South American parasitoid wasp *Microctonus hyperodae* Loan (Hymenoptera: Braconidae) was released in New Zealand in 1991 as a potential biological control agent of the weevil. *M. hyperodae* is a proovigenic species, so its lifetime complement of eggs is set at emergence as an adult from the host. Adult fitness is likely to be affected by host nutrition, because the parasitoid resources carried over from the larval to the pupal and adult stages vary with the size and nutritional quality of the host and the time available for larval development. This research has investigated the effect of tri-trophic level interactions on the parasitoid by manipulating host diet (i.e. pollen availability) and measuring effects on parasitoid fitness (i.e. egg load). This proposed mechanism, whereby the third trophic level may benefit from "resource subsidies" made available to the second trophic level, leads to the suggestion of a new mechanism of enhancing parasitoid fitness- "Indirect Conservation Biocontrol".

Biological control of tephritid flies: the USDA-ARS program in Hawaii & efforts to mitigate fruit fly threats worldwide through international cooperation

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Establishment of melon fly (*Bactrocera cucurbitae*) (1895), Mediterranean fruit fly (*Ceratitis capitata*) (1910), and oriental fruit fly (*Bactrocera dorsalis*) (1945) in Hawaii prompted the introduction of many natural enemies. Initial efforts were undertaken in 1912 by F. Silvestri, who searched primarily for parasitoids of melon and Mediterranean fruit flies. Exploration for additional natural enemies intensified when oriental fruit fly was discovered and a total of 32 natural enemies were released between 1947 and 1952. Today, as a result of these efforts, the egg parasitoid *Fopius arisanus* and the larval parasitoids *F. vandenboschi*, *Psyttalia fletcheri*, *P. incisi*, *Diachasmimorpha longicaudata*, and *D. tryoni* are established. They have played a major role in sustained reduction of fruit flies throughout the Hawaiian Islands. During the past 25 years, investigations have addressed parasitoid ecology, biology, and mass rearing methods. More recently, efforts have addressed better utilization of these species through releases of parasitoids in combination with sterile flies for area-wide suppression of fruit flies, integration of reduced risk insecticides with parasitoids into area-wide IPM programs, and shipments of parasitoids from Hawaii to other areas of the world for classical biological control of fruit flies.

An *Endoreticulatus* species from *Ocinara lida* (Lepidoptera: Bombycidae) in Taiwan

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A microsporidian species is described from the *Ficus* pest, *Ocinara lida*, in Taiwan, The taxonomic position of this species was primitively determined by the soluble protein content of spores and the small subunit rRNA gene sequence (SSUrRNA sequence). Analysis of the SSUrRNA indicated that this isolate from *O. lida* is a member of the genus *Endoreticulatus* and belongs to the microsporidian groups II based on the phylogenetic analysis among microsporidia. The exact taxonomic position of this isolate was confirmed again based on its ultrastructural characteristics of spore and life cycle. The congruent results between SSUrRNA sequence analysis and ultrastructural characteristics show that this isolate is more closely related to *E. bombycis* than *E. schubergi* Zwölfer.

Mass-rearing & sterilisation alter mating behaviour of male Queensland fruit fly

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The effects of domestication and irradiation on mating behaviour of males of *Bactrocera tryoni* (Froggatt) (Diptera: Tephritidae) were investigated by caging wild, laboratory-domesticated and sterile (laboratory-domesticated, gamma-irradiated) males with wild females. Mating strategies of mass-reared males were different from that of wild males, although mating behaviour of wild and sterile males was similar. Mass-reared males were found to engage in mounting of other males much more frequently than wild and sterile males, and began calling earlier. Unnatural selection pressures imposed in mass-rearing conditions may explain changes in mass-reared male behaviour. It is likely that the difference between mass-reared and sterile male behaviour is due to a decrease in sexual motivation and overall mating performance as a result of irradiation. Male calling did not appear to be associated with female choice of mating partners, although this does not exclude the possibility that calling is a cue used by females to discriminate between mating partners. Despite differences in behaviour, frequency of successful copulations and mating success were similar between wild, mass-reared and sterile males, suggesting that wild females in the field may not discern a difference between wild males and sterile male *B. tryoni* released in sterile insect technique programs.

Area-wide biological control of citrus pests in California

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For 80 years, the Fillmore Citrus Protective District (FCPD) has successfully conducted area-wide control of citrus pests. The FCPD was formed in 1922 to eradicate California red scale, *Aonidiella aurantii* , while reducing the risk of infestation to the individual grower. Prior to the formation of the FCPD, 87% of red scale findings in Ventura Co. occurred in this district. By 1928, this was reduced to only 6%. The Fillmore insectary was built in 1926 to rear a predator of the mealybug, *Pseudococcus calceolariae*. The insectary has reared over 25 natural enemies introduced by the University of California during the past 80 years. Black scale was the major pest prior to the introduction of *Metaphycus helvolus* in 1937. Release of *M. helvolus* reduced insecticide applications in the FCPD from 80% of orchards each year to less than 5%. Red scale became the major pest having been partially controlled by the black scale treatments. The FCPD began rearing and releasing *Aphytis melinus* in 1960. In 1986, less than 1% of the FCPD acreage required insecticide treatment. Various pests have threatened the FCPD, but in the end, the collapse of the Valencia market forced the insectary to shut its doors in 2000.

Program for biological control of purple loosestrife in Illinois, USA.

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Since 1994, we have been involved in rearing and distributing *Galerucella* spp. (Coleoptera: Chrysomelidae) beetles for control of purple loosestrife in wetlands in Illinois, USA. Adult beetles reared at the Illinois Natural History Survey have been distributed to cooperators throughout the state, for release at > 230 wetland sites. Success at numerous sites has shown reduction of loosestrife flowering and densities, and many of these sites have shown sufficient regeneration of native flora to keep loosestrife at low densities. We have implemented an education program in which nearly 400 educators have bent rained about wetlands, native biodiversity, invasive species and biological control, using purple loosestrife as a model. Educators have been given all supplies to grow loosestrife and *Galerucella* beetles in classrooms, with the release of beetles by K-12 students into nearby wetlands. Placing biological control into the classroom has led to an increased awareness and acceptance of biological control by the public, which will be necessary as other weeds of natural areas are targeted in the future.

Control of lettuce aphid by the Tasmanian lacewing in New Zealand

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Lettuce aphid (*Nasonovia ribisnigri*) was detected in Christchurch on lettuce in March 2002 and has spread to all lettuce growing regions in New Zealand. The indigenous Tasmanian lacewing, *Micromus tasmaniae*, was an effective predator of this aphid in spring trials (Oct-Dec) at Pukekohe (South Auckland) in 2002–04. In both years aphid populations at harvest in unsprayed plots of lettuces where *Micromus* established were equivalent to or lower than in lettuces to which topical insecticides had been applied weekly. Lettuces from the unsprayed plots were deemed saleable; the mean number of aphids per lettuce was less than 4 in both trials. The mean number of *Micromus* (all stages) peaked at 4.6 and 3.6 per lettuce. Biological control was not maintained in the summer trials (Jan-Mar) when populations of aphids were too high and *Micromus* numbers too low. At harvest the mean numbers of aphids in lettuce from unsprayed plants in the two summer trials were 408 and 121 respectively. Even in treatments where selective insecticides (pirimicarb and pymetrozine) were used, aphid numbers over summer were unacceptably high. *Micromus* is an important predator in a developing IPM programme featuring selective insecticides, imidacloprid drenches, lettuce aphid resistant cultivars, and naturally occurring fungal pathogens for aphid control in field lettuce crops.

Organization & molecular characterization of genes in the polyhedrin region of the *Lymantria xylina* multinucleocapsid NPV

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A multinucleocapsid nucleopolyhedrovirus (MNPV) was isolated from the casuarina moth, *Lymantria xylina*. This virus (LyxyMNPV) has been considered to be closely related to the *L. dispar* MNPV (LdMNPV) based on its *in vitro* host range and protein content. Here, the relatedness of the LyxyMNPV and LdMNPV genomes was examined. Restriction maps with the endonucleases *Bam*HI, *Eco*RI, and *Eco*RV showed significant differences between the two viruses, but most of both DNAs hybridized strongly to each other in reciprocal hybridization experiments. An *Eco*RV-K fragment (4.6 kbp) of LyxyMNPV (polyhedrin gene content region) was compared to the corresponding region of LdMNPV genome, and the results revealed that it is a conservative region in genetic organization. Both polyhedrin genes showed the highest relatedness in their sequence identity (92%) while genes for LdOrf163/-like and LdOrf162/-like peptides (located at the upstream of polyhedrin gene) and essential structural protein and protein kinase (located at the downstream of polyhedrin gene) showed relative low identity. Furthermore, the gene of LyxyNPV LD130-like protein (located outside the LyxyMNPV *Eco*RV-K fragment) was also sequenced, and it showed 93% identity to that of LdMNPV. Therefore, we conclude that LyxyMNPV is a new virus but is a member of the LdMNPV group.

Control of yellowish elongate chafer, ***Hepthophylla picea*** **(Coleoptera: Scarabaeidae) by entomopathogenic fungus,** *Beauveria amorpha*

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Yellowish elongate chafer (YEC), *Heptophylla picea* is the major pest of tea plants, or many kinds of plants in Japan. Especially, larval damage is very serious in tea fields. In general, larvae are controlled using chemical insecticides (fenitrothion). But the effect of fenitrothion is not enough, because larval habitat is a deep in soil at the application time of chemical insecticides. We searched alternative agent of chemical insecticides. The entomopathogenic fungus, *Beauveria amorpha* (Strain: HpBa-1) is the effective control agent to YEC larvae. This fungus has the high virulence to YEC larvae under laboratory and field conditions. YEC larvae have behavioral features, which they move to surface of soil (0~10cm) to consume tea root. This phenomenon appears in early winter, so *B. amorpha* should be applied at this time. Study the dynamics of *B. amorpha* spores at low temperatures (temperature of early winter: 10 °C) *in vivo* and *in vitro* is necessary to control YEC larvae using *B. amorpha* effectively. In this study, we investigated ecology and effects (virulence, inhibition of food consumption) of *B. amorpha* at low temperatures. Furthermore, we suggest the appropriate time of application of this fungus.

Isolation of a highly toxic strain of *Bacillus thuringiensis* & its effects on the growth & development of *Spodoptera exigua* (Lepidoptera: Noctuidae)

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In this study, a strain of *Bacillus thuringiensis* YL-17 highly toxic to *Spodoptera exigua* was isolated from 23 stock *B. thuringiensis* isolates. Its lethal and sublethal effects on *S. exigua* were evaluated through investigating 50% lethal concentration (LC_{50}) values and development and mortality in response to sublethal doses of *B. thuringiensis*. The LC_{50}s of pure crystals, pure spores and the spore-crystal complex were 22.1μg/ml, 230.2μg/ml and 20.2μg/ml, respectively. The toxicity of *B. thuringiensis* YL-17 mainly results from its crystal. When it applied to eggs, *B. thuringiensis* YL-17 did not affect the hatch rate, but the neonate mortality increased significantly ($P<0.05$) with an increase of treated concentration and egg age. Ingestion of the spore-crystal complex significantly ($P<0.05$) inhibited the growth of third instar larvae, significantly ($P<0.05$) extended the duration of larval development, and caused a significant ($P<0.05$) reduction in the weight of pupae, the pupation rates and the adult emergence rates of larvae that survived. There was no significant effect ($P<0.05$) on adult sex ratio among the survivors. These results show that *B. thuringiensis* YL-17 not only has a direct effect, causing death of *S. exigua* larvae, but at lower doses also produces sub-lethal detrimental effects on larvae and pupae.

Pathogenicity of *Metarhizium flavoviride* to rice planthoppers

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The susceptibility and virulence of entomopathogenic fungus *Metaarhizium flavoviride* to the developmental stages of brown planthopper (BPH), *Nilaparvata lugens* (Stal) and whitebacked planthopper (WBPH), *Sogatella furcifera* (Horvath), were investigated under laboratory conditions. Three concentrations of *M. flavoviride* ranging from 10^6 to 10^8 conidial/ml were included in the assay. The tested stages of hosts were small nymphs (1-2 instars), large nymphs (3-5 instars) and adults. It was shown that both stages of hosts were susceptible to the fungal infection. The BPH and WBPH infection by *M. flavoviride* were observed mostly on days from 1 to 21 after exposure. All the planthoppers killed sporulated well and displayed typical signs of *M. flavoviride* infection under microscopic examination after overnight incubation in the moist chamber. The adults were more susceptible to infect *M. flavoviride* than their nymphs and the small nymphs were most resistant to fungal infection. The cumulative mortality at all stages was concentrations-dependent.

Key words: Planthopper, *Metarhizium flavoviride*, susceptibility, virulence

A method to trap and rapidly segregate subterranean termites (Isoptera: *Rhinotermitidae*) for laboratory bioassays

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A simple technique to trap and rapidly segregate large number of subterranean termite (*Rhinotermitidae*) using bamboo culms is described. The technique separates termites from their natural substrates by placing moist, weathered bamboo culms on top of broken nest debris. Termites immediately migrate from their natural substrates to the dark and moist cavities of the culms allowing rapid collection by tapping out one end onto flat trays when sufficient numbers of termites aggregate in the interior cavities. Field trapping technique involves the use of bundles of weathered, moist bamboo culms placed horizontally along foundation walls or strategic locations in a known area infested with termites. Termites readily feed on the nutrient rich bamboo, aggregate inside the cavities while feeding and then collected by tapping out one end onto flat trays. These techniques provide a fast, inexpensive and efficient method of trapping and segregating tens of thousand of live termites with no or minimal injury.

Killing insects in malting barley and canola with heat – are the seeds alright?

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Serious problems confront chemical disinfestation of insects in stored products. Methyl bromide is becoming costly and non-quarantine usage phased out. Phosphine is increasingly regulated and insect resistance is growing. Resistance to protectants is also common and pesticide residues are more unacceptable to markets. In comparison, heat disinfestation has the advantages of being chemical and resistance free, and is also rapid. Extensive work has demonstrated that the most heat tolerant of stored product insects can be successfully removed from large tonnages of wheat without any adverse effects on the grain. There is considerable value in expanding the use of this physical method to the disinfestation of malting barley and canola. However, there are vital quality parameters of the commodities that may potentially be heat sensitive. This poster summaries results of germination tests on the two seed types, barley micro-malting and canola oil quality analysis, for a range of heat treatments required to disinfest *Rhyzopertha dominica* and *Trogoderma variabile* at temperatures from 50ºC to 60ºC. The implications of the results for the feasibility of using heat disinfestation are outlined.

Rapid degradation of termiticides under field conditions

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In Australia, some state regulatory agencies may collect soil samples to determine the adequacy of termiticide application by pest controllers. Soil tests may be carried out months or even years after treatment, making an accurate knowledge of termiticide degradation rates crucial if determinations are to be valid. Termiticide degradation was investigated as part of a field trial in southern NSW. Termiticides were mixed with soil at label rates to a depth of 700 mm. One day and one year after treatment samples were collected to a depth of 75 mm. Mean losses at 12 months were: chlorfenapyr 24%, bifenthrin 30%, imidacloprid 94%, fipronil 96%, chlorpyrifos 99%. Fifteen months after treatment, plots treated with chlorpyrifos, fipronil and imidacloprid were sampled again, to a depth of 450 mm in 150 mm increments. In soil below 150 mm, chlorpyrifos and fipronil content was little changed from time of application whereas major losses of imidacloprid had occurred throughout the soil profile. These results have implications for termite treatment regulation in Australia. Regulatory agencies have relied upon degradation rates observed in laboratory experiments to determine *in situ* treatment adequacy. My field study results suggest that termiticides can degrade more rapidly than these studies have indicated.

Laboratory and field trials with juvenogens as potential termite control agents

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Juvenogens are complex compounds from which the active part of the juvenile hormone (JH) mimicking molecule is liberated by metabolic processes within the insect body. Eight juvenogens were prepared from parent juvenoid alcohols. They showed a high juvenilizing effect in tests with the termite *Prorhinotermes simplex.* This effect is manifested by differentiation of termite workers into presoldiers, soldiers or soldier/worker-intercastes. *cis*-Ethyl *N*-{2-{4-[2-butanoyloxycyclohexyl)methyl]phenoxy}ethyl}carbamate was tested with *Zootermopsis angusticollis, Kalotermes flavicollis, Cryptotermes declivis, Reticulitermes santonensis*, and *R. flaviceps* and proved efficacious in force-feeding (FF) and in choice bioassays. After application of 0.5 mg ml^{-1} in FF experiments with the two economically important species of *Reticulitermes,* approx. 57% resp.73% of workers changed into soldiers. In *Z. angusticollis* application of 0.05 mg ml^{-1} caused differentiation in 62% soldiers; 57% soldiers differentiated after topical application of 1mg of compound per termite. Differentiation of excessive soldiers induced by juvenogens may cause disruption of the social structure and ultimately the death of the colony, a concept that is currently being evaluated in the field with Australian species of termite. Juvenogens are promising candidates as actives in bait technology for control of subterranean termites. Financial support of the Grant Agency of the Academy of Sciences of the Czech Republic (grant No. S4055104) is gratefully acknowledged.

The effect of amylase inhibitor on target and non-target stored-product mites

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A combination of biological control and biochemical control of stored-product mite was evaluated in laboratory conditions. The inhibitor of digestive enzyme amylase was applied as a biochemical acaricide. The amylase inhibitor had a potential to suppress the population of pest flour mite (*Acarus siro*), however, did not lead to extinction of the flour mite. Predatory mite (*Cheyletus malaccensis*) is a predator of flour mites in granary habitats, which is used in biological control of the pest mites as an alternative or complement to application of pesticides in postharvest protection. The suppressive effect of the amylase inhibitor on the target flour mite and the non-target predatory mite was tested *in vitro* and in *vivo*. The experiments *in vitro* indicated a high enzymatic activity of digestive amylases and low enzymatic activity of proteinases in flour mites as an evolutionary adaptation to starch being the main energy source. Oppositely, the predatory mites had high content of proteinases and low content of amylases indicating an intensive digestion of proteins from animal tissues. The amylase inhibitor was found more effective against the flour mite amylases than the predatory mite amylases. The experiments *in vivo* were performed on diet enriched by the amylase inhibitor in a trophic system with *A. siro* and *C. malaccensis*. In comparison to control condition, the applied level of the amylase inhibitor reduced population growth of *A. siro* to 35 %. No adverse effect of the amylase inhibitor on predator longevity and the length of oviposition period was observed. The prey reared on the inhibitory diet influenced negatively the females of *C. malaccensis* as they produced less eggs than those reared on the control prey. The lower energy content of prey (namely its weight) grown on the inhibitory diet was compensated by higher predatory rate of *C. malaccensis*. The results show that the biochemical acaricides are good candidates for regulation of stored-product mites in combination with biological approach. The data are discussed in the context of using of GMO methods in the integrated control of mite pests in post-harvest protection. *The project was supported by grant COST OC 842.20 and GACR 203/02/P081*

Movement of adult *Cryptolestes ferrugineus* (Coleoptera: Cucujidae) in stored wheat in response to temperature gradients and moisture differences

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The movement and distribution of adult *Cryptolestes ferrugineus* (Stephens) (Coleoptera: Cucujidae) were determined in 100×100×1000 mm wheat columns with or without 5 C/m temperature gradients (from 27.5±0.2 to 32.5±0.2 C or at 27.5±0.2 C) under half of the columns with 12.5% or 14.5% m.c. wheat and the other half with 16.5% m.c. wheat. Adults introduced in the middle of the horizontal wheat columns with uniform temperature showed no bias in the direction of net displacement, with the distribution pattern gradually becoming more uniform when time increased from 1 to 144 h. Adults responded to both temperature gradients and moisture differences in columns of wheat with temperature gradients and moisture content differences. The response was different at different moisture conditions. Adults were more sensitive to moisture differences and moved faster in 12.5% m.c. wheat than in 14.5% m.c. wheat. Adults stayed in warmer sections in both high and low moisture regions. At different moisture conditions, adults changed their preference (e.g., preferred high moisture grain in dry grain, and preferred warmer temperature in wet grain).

Essential oils as fumigants in stored grain protection

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Essential oils from croatian aromatic plants: lavander, laurel, rosemary and thymus, and their single components had contact and fumigant activity to the major stored grain pests resistant to the phosphine-based fumigants applied so far. By gas-chromatography nine most significant oil components were determined: 1,8-cineole, camphor, eugenol, linalool, carvacrol, thymol, borneol, bornyl-acetate, linalyl-acetate, and applied to the wheat and corn samples infested with seven pest species: *S. granarius*, *S. oryzae*, *S. zeamais*, *R. dominica, T. castaneum, O. surinamensis,* and *C. ferrugineus*. Laboratory and storages analyses showed that highly susceptible primary species to the nine isolates were genus *Sitophilus*, followed by *R. dominica*, when treated with camphor, linalool, and 1,8-cineole, and secondary species *O. surinamensis* when treated with 1,8-cineole, camphor, linalool, and thymol. *C. ferrugineus* was susceptible in treatments with 1,8-cineole, eugenol, camphor, and carvacrol. The exception was *T. castaneum* where application of plant preparations appeared to be insignificant. The most effective component and carrier of fumigant and contact toxicity was 1,8-cineole, followed by camphor, eugenol, carvacrol, linalool, and thymol. Borneol, bornyl-acetate, and linalyl-acetate were less effective. All the components tested showed no negative influence on reological properties of wheat flour.

Spatial orientation of the mould mite in the computer-programmed olfactory field

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The attractiveness of food odors to the mould mite, *Tyrophagus putrescentiae* (Schrank) (Acarina: Acaridae) was demonstrated in a micro-locomotion compensator (MLC) as well as in a classical choice-chamber. The MLC has been developed for use in experiments on the orientation mechanisms of tiny animals. The positioning system of the MLC constantly returns a mite walking on a glass plate to the central position by sliding the plate. While driving the MLC, the main computer switches a micro solenoid-valve to generate odor cues in airflow over the mite. If the odor is generated by position in a virtual olfactory field created by the MLC, the mite would exert a similar olfactory response as in a real choice-chamber. In practice, odorous zones with attractant were defined on the glass plate in a checkered pattern, and the test animal was exposed to the odor while it stayed in the zones. The frequency of stays in the odorous zones positively correlated with the odor concentration to provide a reliable estimate of the attractiveness using probit analysis.

Control of onion thrips (*Thrips tabaci*) in stored red onions with pyrethrum insecticide

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The onion thrip, *Thrips tabaci*, is a worldwide pest of onion crops. In the 2001/2002 season, damage caused by onion thrips feeding on red onions stored in Tasmania led to more than 50% of onions being lowered in quality and value. This study examined the efficacy of PestigasTM, on juvenile and adult *Thrips tabaci* in laboratory trials and in onions in simulated storage conditions. PestigasTM is a high pressure aerosol insecticide containing natural pyrethrum and piperonyl butoxide. In laboratory trials, mortality was significantly higher in adults than juveniles, with 72% and 54% mortality respectively. Increasing the rate of PestigasTM from 150g/300m3 to 1.2 kg/300m3 increased mortality of thrips, with significantly higher thrips mortality (i.e. 81%) when applied at 1.2kg/300m3. In simulated storage trials, significantly less larvae (i.e. first and second instar) and adults were alive in onions treated with a single application of PestigasTM at 1.2kg/300m3. The largest PestigasTM treatment effect occurred with adult thrips where treatment reduced the average count from 6.8 adults in untreated onions to 1.8 adults in PestigasTM treated onions. Repeated applications of PestigasTM at 1.2kg/300m3 reduced the number of live thrips from 70-100% in comparison to untreated onions. Further trials investigating the effect of repeated applications of PestigasTM on onion quality and thrips survival are being undertaken.

Study on biology of *Lasioderma serricorne* F. under laboratory conditions

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Two series experiments were carried out on the biology of tobacco beetle, *Lasioderma serricorne* F. (Col.: Anobiidae) a destructive pest in tobacco warehouses in Iran at constant temperature 29±1oC and 70-75٪RH and in the variable temperature ranging between 20-27 and 14-65٪ RH in Guilan University. There were highly significant differences among all developmental stages at two mentioned conditions. The average developmental time of a generation was 60.69±1.59 and 127.58±1.35 days at constant and variable temperature and humidity, respectively. The percentage of mortality in all stages was higher in variable condition than mortality in constant temperature except, incubation period that was higher at constant temperature. Temperature and humidity affect on longevity of tobacco beetle and females lived longer than males in both conditions. Most oviposition occurred within 5 days of females emergence. Mean number of eggs laid by a female was 73.17±6.76 and 78.47±4.65 in variable and constant conditions, respectively.

Susceptibility of some dried medicinal plants to the attack of *Oryzaephilus surinamensis* (L.) (Coleoptera: Silvanidae)

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The susceptibility of 24 dried medicinal plants commonly used in food industry to the attack of *Oryzaephilus surinamensis* (L) was valued. 50 eggs, with 4 replications for each test carried out at 26 ± 1°C and 70 ± 5% r.h. were placed on 20g of dried vegetal. The development of the beetle was observed on *Angelica archangelica*, *Citrus aurantium*, *Crataegus oxyacantha*, *Foeniculum vulgare*, *Juniperus communis*, *Matricaria chamomilla*, *Rheum officinale*, *Silybum marianum*, *Valeriana officinalis* and *Vitis vinifera*. The highest mean number of adults (42.0 ± 2.16), the shortest mean period of development (35.0 ± 0.00 days) and the highest mean index of susceptibility (4.6 ± 0.06 %) were observed on *Silybum marianum*. The lowest mean number of adults (2.5 ± 0.58), the longest mean period of development (84.5 ± 1.29 days) and the lowest mean index of susceptibility (0.5 ± 0.12 %) were observed on *Matricaria chamomilla*. Various factors affected the insect development on 14 medicinal plants: quality and quantity of nourishment, presence of repellent, antifeedant active ingredients or with insecticide action, besides for the substrates structure (shape, dimension and state).

Program participants' knowledge and perceptions of effectiveness of the French Quarter Formosan Termite program

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Under Operation Fullstop, the LSU Agricultural Center, in cooperation with the Agricultural Research Service, began a large area pilot test in the New Orleans French Quarter. The program was designed to demonstrate the effectiveness of area-wide management of pests like the Formosan subterranean termite. Four years after the beginning of the program, data indicated that the population of the Formosan termite in the original 15-block area of the French Quarter program was being reduced, indicating to LSU Agricultural Center and ARS personnel that the program was working. It became evident, however, that the French Quarter program needed to be evaluated from the viewpoint of the participants of the study in order to assess their perceptions of the effectiveness of the program and to measure their knowledge of educational aspects of the program. Based on results of the study, it was concluded that the majority of the participants thought that the program was important, effective, and should be continued and expanded to other areas of the French Quarter. Knowledge scores derived from specific survey questions indicated that the participants were knowledgeable about termite biology, identification, prevention, control, and management.

A survey of natural enemies on stored-rice insect pests in Thailand

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Although rice is the most important food crop in Asia, losses of stored rice due to insect infestation in the ASEAN countries are tremendously high during storage and distribution, under hot and humid climatic conditions. Biological control by using natural enemies is one of the effective methods to control stored-product insect pests. For improvement in the use of these natural enemies, it is necessary to clarify the fauna of natural enemies of stored-rice insect pests. Thus, we surveyed parasitoids and predators of stored rice pests all over Thailand. As the results, 29 species of hymenopterous parasitoids of 7 families and 53 species of predators belonging to 5 orders were collected from rice stores in Thailand. Some parasitoids (*Proconura minusa*, *Theocolax elegans*, *Cerocephala dinoderi*, *Anisopteromalus calandrae*, *Lariophagus distinguendus* and *Bracon hebetor*) and some other hemipteran species (*Amphibolus venator*, *Peregrinator biannulipe* and *Xylocoris flavipes*) were most abundant all over Thailand, and were estimated as potential biological control agents for stored-rice insect pests.

Water dependence of Japanese subterranean termites, *Coptotermes formosanus* **Shiraki and** *Reticulitermes speratus* **(Kolbe)**

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In Japan, *Coptotermes formosanus* Shiraki and *Reticulitermes speratus* (Kolbe) are economically major species. The purpose of this study is to determine the relationship between the feeding activities of these Japanese subterranean termites, and their environmental conditions in terms of water. First, relationships between desiccation and physiological damages to both species were evaluated. Among 9 combinations (20, 25, and 30°C-50, 70, 90% relative humidity (RH)) of the desiccating condition (without a water supply), the longest and shortest survival periods of both species were obtained under the 20°C-90% RH and 30°C-50% RH conditions, respectively. Comparing to tolerances of mass loss for recovery from the damages caused by desiccating condition, higher body mass was measured in *R. speratus*. Secondly, effects of RH, temperature, and moisture content (MC) of wood on the feeding activities of termites are evaluated under natural condition (with a water supply). The optimum temperature-RH combinations for feeding activities were 30°C-90% RH for *C. formosanus*, and around 30°C and 70-90% RH for *R. speratus*. In addition, feeding preferences were found in the middle MC (79-103%) blocks for *C. formosanus*, and the middle and high MC (140-182%) blocks for *R. speratus*, whereas even wood blocks of low MC (6-12%) were attacked.

An environmentally friendly alternative to phosphine gas

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The Australian grain industry is worth about $7 billion per annum, with approximately 80% of production exported. To maintain a competitive edge, the Australian grain industry needs to market a predictably high quality product. There are serious problems caused by both introduced and indigenous insect pests of storage grain, and by the fact that these insect pests have developed resistance to commercial fumigants. Fumigation with phosphine gas is the key component in the management of insect pests in stored grains. These stored-grain pests in Australia now exhibit resistance to phosphine at levels more than 200 times the original lethal dose. Therefore the industry is searching for alternatives. In this study we have tested different concentrations of eucalyptus oil and its major component, 1,8-cineole against rust-flour beetle (*Tribolium castaneum*) and lesser grain borer (*Rhizopertha dominica*). The results were evaluated against the discriminating dose of the commercial fumigant -phosphine gas. The preliminary results indicate that 1,8-cineole could be useful in developing an environmentally friendly fumigant to control stored grain pests.

Distribution of the warehouse beetle *Trogoderma variabile* (Coleoptera : Dermestidae) as a pest of bulk grain storage in Australia

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The warehouse beetle *Trogoderma variabile* was first detected in Australia in the late 1970s, in southern New South Wales (NSW). Despite several attempts at eradication, *T. variabile* became a persistent and hard- to- eliminate pest of storage structures and a pest of bulk-stored canola. To update knowledge on its distribution a survey was undertaken. During the summers of 2001-02 and 2002-03 pheromone-baited sticky flight traps were deployed around grain storage and handling facilities on the Australian mainland. Results indicate that *T. variabile* is now widespread through the south eastern 'wheat belt', from northern NSW to eastern South Australia (SA), but remains uncommon in Queensland (Qld.) and Western Australia (WA). Of grain growing regions, only the Eyre Peninsula of western SA appears free of this pest. Comparison with 1990-93 data shows that *T. variabile* has consolidated its position in south eastern Australian 'wheat belt' where it is now effectively ubiquitous. Spread of *T. variabile* into Qld. may be limited by climate. Its apparent rarity in WA is hard to explain, but may be partly due to the lower densities of suitable habitats (human settlements and poorly run 'private' and on-farm storage) making successful dispersal to new sites more difficult and slower.

Area-wide pest management of Formosan subterranean termites in the French Quarter, New Orleans, Louisiana, USA

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The Formosan subterranean termite, *Coptotermes formosanus*, is a serious pest where it has become established and is one of the most destructive insects in Louisiana. A pilot test was begun in 1998 in the French Quarter to demonstrate the effectiveness of using area wide management. All properties in a contiguous 15 block area were treated using commercially available baits or non repellent termiticides. In 2002 the treatment zone was expanded. Glue boards were used to estimate alate numbers and in-ground monitors for foraging activity. Alates were sampled once a week in April and two to three times weekly from May to July 15 in 1998 through 2003. Monthly monitoring of foraging activity began in January, 1999. These data showed that area-wide management reduced termite activity by 50 %. Isolated areas of "high" termite activity remain inside the test area. Inspections of properties using infrared and acoustic detection technologies are being conducted to allow discovery and treatment of colonies which have, so far, escaped treatment. A third expansion began in March of 2003. Continued treatment, expansion, and monitoring are required to assess the long-term effects of the area wide management program.

Amylase inhibitors as protein biopesticides

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The amylase inhibitors represent a perspective protectant of stored-food commodities against pest arthropods. We review current knowledge on amylase inhibitors of plant origin, and present the experimental work on their efficacy against the selected storage insect and mite pests. Six structural classes of plant amylase inhibitors of protein character were characterized on molecular level including Lectin, Knottin, Cereal, Kunitz, Thaumatin and γ-Purothionin types. The research on amylase inhibitors encompasses three levels with decreasing frequency of published reports (a) *in vitro* inhibition of insect amylases (53 reports), (b) *in vivo* data on increasing mortality or weight losses of insects (12 reports), (c) application in GM-plants (2 reports). There are no reports available on effect of the amylase inhibitors on storage grain mite pests. We present here both *in vitro* and *in vivo* analysis of suppression of amylase activity in *Acarus siro* as a model mite species. Systematic research is necessary to select the amylase inhibitors with defined propertied and to modify their characteristics by genetic engineering. The biochemical screening correlated with standardized biotests will intensify application of these biopepticides in the crop biotechnology industry. The project was supported by grants GACR 522/04/1286, GACR 203/02/P081, MSMT OC D16.001 and MSMT P1OC 853.003

Development of experimental procedure for determining material susceptibility to termite attack

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As part of an ongoing project investigating risk-based interactions of termites and protective barrier systems, I developed a laboratory methodology for studying the interaction of termites and barrier materials. The test materials are sheets of polyurethane or other synthetic formulations up to 3mm in thickness. Test material is clamped between two glass tubes, one of which leads to a home container housing termites and the other to a food resource. The assay system allows the close observation of termite behavior by using agar and other transparent media as foraging substrates within the glass tubes. Termites from the home container typically tunnel through the foraging substrate and come into contact with the tested material within 24 hours of introduction. The tenacity of termite attack is increased if the surface of the material is scarified rather than shiny. Small breaches in the form of 0.5 mm holes also intensify and focus termite damage. Both *Coptotermes acinaciformis* and *Shedorhinotermes seclusus* have been able to breach high density polyethylene barriers when used in this assay. The assay will be used to determine desirable physical and chemical characteristics of synthetic barrier materials for protection from termite attack under laboratory conditions.

A theoretical approach to evaluate the performance of gravel physical barriers against termites

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The performance of gravel physical barriers against termites is generally described that the gap between gravel particles is too small to the termite penetration, and that even the single particle is too heavy to be removed by termites. However, no theoretical approach such as the arrangement of mandibles and maxillaes of target termite species has been conducted to date. The maximum openings of mandibles and maxillaes of workers of a Japanese major pest species *Coptotermes formosanus* Shiraki were 0.45 mm and 0.88 mm, respectively. In addition, the minimum gap between mandibles and maxillaes was 0.07 mm. This theoretically results in that C. *formosanus* workers can remove particles with less than 1.9 mm diameters if the particle is spherical in shape. Since the maximum head width of *C. formosanus* workers is approximately 1.2 mm, particles with less than 2.9 mm diameters are small enough to prevent termite invasion. These results well coincide with the laboratory test results that many kinds of gravel materials with 1.7 – 2.0 mm diameter have good performance against workers of *C. formosanus*, and that the workers allowed to penetrate into glass beads with >2.8 mm diameter.

Thaumetopoea Processionea (LINNAEUS), a serious problem in densely populated areas (Lepidoptera: Notodontidae)

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Thaumetopoea processionea has become a serious problem at many localities in The Netherlands, Belgium and Germany since its rediscovery in 1987. After more than a century the oak processionary caterpillar (*Thaumetopoea processionea* Linnaeus) re-entered the northwestern parts of Europe. Although definitely not a common species in The Netherlands and Belgium, it was occasionally observed locally in high numbers during the nineteenth century. The main host plant of the oak processionary caterpillar in The Netherlands, Belgium and Germany is common oak (*Quercus robur* Linnaeus). Caterpillars of a number of moth species may present danger to man. In the processionary caterpillar the hazards are caused by the many thousands of urticating micro-hairs developing on the larvae from the third larval stage onwards. The hairs are easily released by touching the caterpillars or their nests and they can float on air over longer distances. After penetrating the skin, the urticating hairs can cause miniscule and painfull wounds. Since 1997 the population of *T. processionea* in The Netherlands as well as in Belgium decreased however, since 2000 the population is on the increase again in many areas. This time even more in a northerly direction.

Tennessee and Kentucky Stored Grain Project: An Update on Progress

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A cooperative effort with the University of Kentucky and the University of Tennessee was established in 2001 to determine if basic procedures and the use of existing pesticides labeled to control insects in wheat would be sufficient to prevent damage from stored grain insects in farm bins in each state. To this date there has been progress in reducing insect problems in demonstration grain bins using these established methods of insect control. This project is funded by an IPM grant to purchase necessary materials and monitoring equipment needed to complete the project.

On Farm Research trials on evaluation of pulse protectants against pulse beetle, *CALLOSOBRUCHUS MACULATUS* (F.) infesting gram , *CICER ARIETINUM* (L.)

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Farm trials of groundnut oil 3.75 ml/kg + turmeric powder 1.75 g/ kg , mustard oil 3.75ml/ kg **+ t**urmeric powder1.75 g/ kg , *neem* oil 10ml/kg and 7 cm sand covering on gram were evaluated against *C. maculatus* on egg basis, adult emergence and damaged grain in treated and untreated gram after 60,120 and 180 days storage. Pulse protectants significantly protected treated gram from damage as compared to untreated after 60,120 and 180 days .After 60 days, 0.0 % damage was recorded in gram treated with groundnut oil 3.75 ml/kg + turmeric powder 1.75 g/kg , mustard oil 7.5 ml/kg + turmeric powder 1.75 g/kg , 7 cm sand covering and *neem* oil 10ml/kg as compared to 6.1% damage in untreated .After 120 and 180 days storage , similar results were observed on gram treated with protectants except in untreated gram wherein damage 21.92 and 46.40 % was recorded, respectively. Groundnut oil 3.75 ml/kg + turmeric powder 1.75 g/kg, mustard oil 7.5 ml/kg + turmeric powder 1.75 g/kg, 7 cm sand covering on gram and *neem* oil 10ml/kg proved effective in protecting treated gram from beetle for 180 days. 43 Demonstrations on mixing pulse protectants were shown in households.

The Bioconversion of Putrescent Waste

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Imagine a bioconversion process that reduces the weight and volume of food waste by over 95% within a matter of just a few hours. The process requires no energy, no electricity, no chemicals, not even water. The process is housed within a container that resembles a small plastic garbage bin. The unit has no moving parts and requires no maintenance. Since it must be emptied once each year or two, it eliminates altogether the collection, transport and land-filling of food waste. The process generates very little odor, and at the same time, it very effectively repels houseflies and other filth-bearing flies. The unit requires very little expertise or experience to operate, and it can handle the daily food waste of more than 25 people. The process is driven by the larvae of the black soldier fly, *Hermetia illucens*, an insect that poses no threat to humans and has never been associated in any way with the transmission of disease. Upon reaching maturity, the larvae migrate out of the unit and into a collection bucket without any human or mechanical intervention. This self-harvesting grub represents a bundle of nutrients that rivals in commercial value the finest fish meal.

Susceptibility of some dried medicinal plants to the attack of *Oryzaephilus surinamensis* (L.) (Coleoptera: Silvanidae)

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The susceptibility of 24 dried medicinal plants commonly used in food industry to the attack of *Oryzaephilus surinamensis* (L) was valued. 50 eggs, with 4 replications for each test carried out at 26 ± 1°C and 70 ± 5% r.h. were placed on 20g of dried vegetal. The development of the beetle was observed on *Angelica archangelica, Citrus aurantium, Crataegus oxyacantha, Foeniculum vulgare, Juniperus communis, Matricaria chamomilla, Rheum officinale, Silybum marianum, Valeriana officinalis* and *Vitis vinifera*. The highest mean number of adults (42.0 ± 2.16), the shortest mean period of development (35.0 ± 0.00 days) and the highest mean index of susceptibility (4.6 ± 0.06 %) were observed on *Silybum marianum*. The lowest mean number of adults (2.5 ± 0.58), the longest mean period of development (84.5 ± 1.29 days) and the lowest mean index of susceptibility (0.5 ± 0.12 %) were observed on *Matricaria chamomilla*. Various factors affected the insect development on 14 medicinal plants: quality and quantity of nourishment, presence of repellent, antifeedant active ingredients or with insecticide action, besides for the substrates structure (shape, dimension and state).

Dispersal, dispersion and trap catch of storage insects in buildings

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Traps are widely used for monitoring storage pests, because they are sensitive indicators of infestation and often yield abundant data for analysis, but biological interpretation of trap catch can be difficult. Accordingly, the objectives of trapping in a particular situation should be carefully defined, because the objectives will determine how the data should be analyzed and interpreted. The main purpose of trapping in buildings (warehouses, processing plants, retail stores, etc.), aside from insect detection, is to identify foci of infestation and points of entry. Thus, interpretation of trap catch in buildings focuses on population distribution (dispersion) rather than population density. The number of insects captured by any trap is determined by the frequency of insect-trap encounters and trap efficiency. Frequency of encounter depends upon numbers dispersing (from foci of infestation and entry points), the attraction of the trap, and trap location (relative to foci of infestation, entry points and pathways of dispersal). Pathways of dispersal and their influence on dispersion of trap catch are examined, and their importance in trap catch interpretation is discussed.

New innovations and trends for insect pest management in stored and processed food

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There is increased interest in using integrated controls to manage insects in stored products, which include a continuum from raw grain to processed food products. However, integrated pest management (IPM) in stored products, particularly in stored and processed food, often involves multiple components that may or may not use sampling as part of the decision-making process. This is in sharp contrast to classical IPM as developed for managing insects in field crops, partly because of the consequences of insect infestation in finished or packaged food products. Examples of components used in an IPM program for stored processed food include but are not limited to reduced risk-insecticides, cleaning and sanitation, pheromone monitoring to track population dispersal, lethal temperatures and heat treatments, and spatial mapping for targeted pesticide application. Selected examples from recent research will be used to illustrate concepts and ideas. Future trends and potential impacts on research will also be described and discussed.

Eavesdropping on marsupial moles and their invertebrate prey

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Obtaining information on the lifestyles and habits of subterranean animals is often a great challenge. Marsupial moles (*Notoryctes* spp.), are among the most elusive and least known vertebrates in Australia, and are currently regarded as Endangered. The most fossorial of marsupials, *Notoryctes* live in the vast arid sandy deserts of Central Australia. They are insectivorous, apparently specializing on underground colonial insects such as ants and termites, and very rarely venture to the surface. Underground they use their spade-like hands to tunnel through the sand and backfill as they move along. Using geophones, we have recorded the tunneling sounds of *Notoryctes* that were caught on the surface and released, and these signature sounds appear distinctive and unique. In order to learn about marsupial mole ranging behaviour, activity cycles and abundance we have developed a system to track an animal's movements underground using a grid of geophones. The system records from up to 32 geophones simultaneously over an area of about 0.3 ha, and operates on an adjustable recording schedule. While the system has been designed to elucidate aspects of the conservation ecology of marsupial moles, we also hope to be able to plot, dig up and identify invertebrates by their signals.

Mapping weevil and larger grain borer resistance and putative resistance factors in tropical maize.

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The maize weevil, *Sitophilus zeamais* (Motsch.) and larger grain borer, *Prostephanus truncatus* (Horn) are important pests of stored maize (*Zea mays,* L.), which cause severe losses in tropical agroecologies. The International Maize and Wheat Improvement Center has utilized germplasm bank accessions to develop sources of resistance as well as a molecular map to quantitative trait loci (QTL) associated with storage pest resistance in a tropical maize (Population 28). Resistance to the maize weevil was evaluated using four susceptibility parameters under controlled condition. Using composite interval mapping (CIM), five QTL on chromosomes 4, 5, 6 (2 QTL), and 9 explained 24% of the phenotypic variance (σ^2_p) for grain damage. Eight QTL explained only 22.5% of σ^2_p for grain weight loss. Nine QTL (31% of σ^2_p) were identified for the Dobie Index. Four main areas for resistance were identified in bin 1.05, 6.00/1, 6.05, and 9.05, with 6.01 accounting for the most phenotypic variation. These regions map in areas know to carry genes associated with cell wall biochemistry that may be considered candidate genes for using in marker assisted selection or genetic engineering.

Efficacy of spinosad and chlorpyrifos-methyl for controlling insects infesting stored wheat

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Laboratory and field experiments were conducted to compare the efficacy of spinosad, chlorpyrifos-methyl, and a combination of spinosad and chlorpyrifos-methyl as grain protectants on stored wheat. Bioassays for adult mortality and progeny production were conducted at 28 C and ~65% relative humidity. Any treatment that contained spinosad at 1 ppm or greater effectively controlled adult *Rhyzopertha dominica* and prevented progeny development in both laboratory and field studies for 48 weeks. Spinosad alone and at higher combinations effectively controlled *Sitophilus oryzae* adults in laboratory studies but did not prevent progeny production probably because eggs are laid inside kernels where larvae are protected from pesticide exposure. Spinosad effectiveness against *Tribolium castaneum* adults decreased through time with few progeny produced. *Oryzaephilus surinamensis* adults were poorly controlled at the lower treatment levels. Although over 90% of *Plodia interpunctella* eggs hatched, no surviving larvae were found in any insecticide treatment through 36 weeks whereas >92% of the eclosed eggs were recovered as larvae from controls. Overall, combining chlorpyrifos-methyl with spinosad did not improve efficacy, and in some cases the combination decreased the effectiveness of spinosad. Spinosad will be an effective alternative insecticide against *R. dominica* when it receives registration for use on stored grains.

Cockroach bait aversion and the development of a new gel bait formula

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Cockroach baits are the most effective and widely used formulation for cockroach management in the United States. Among bait types, gel baits are the preferred formulation of pest management professionals. For many professionals gel baits have totally replaced residual sprays, dusts, aerosols and other cockroach treatments. Baits continue to perform well in the majority of sites; however, in an estimated 5% of accounts in the U.S. baits are not providing the high levels of control professionals have come to expect. Cockroach gel bait aversion occurs as cockroaches develop a feeding aversion to the food ingredients in baits. This characteristic is passed on to offspring and has resulted in reduced bait acceptance and poor control. Bait aversion is not new. Aversion to gel baits by the German cockroach, *Blattela germanica* was first reported in 1990 by Maxforce researchers working with professionals in Orlando, FL. After considerable research it was discovered that cockroaches had developed an aversion to glucose, a major component in gel baits. Glucose was replaced with fructose in a new Maxforce gel bait formula and once again cockroaches were highly attracted to the bait. During the summer of 1999, professionals again discovered bait averse cockroaches. Maxforce researchers (now Bayer Environmental Science) began work to better understand the problem and to make further improvements to the Maxforce bait formula. Suspected averse cockroach stains were collected by professionals from sites across the U.S. and sent to Bayer's research laboratory in Clayton, NC. Existing Maxforce bait formulas and competitive gel baits were tested against the averse strains. All baits tested were less effective against the averse strains than susceptible laboratory strains. Next, the Maxforce bait formulas were dissected to determine the component(s) that were responsible for the aversion. Greater than 50 new bait formulas were tested for bait consumption and mortality against averse strains. Ultimately, a final formula was selected for commercial development. Field testing with professionals confirmed the new formula's efficacy in controlling averse cockroach strains. Bayer Environmental Science launched Maxforce^{Ò} FC Select Roach Killer Bait Gel in the spring of 2004 for use by professionals in the U.S. This presentation will provide basic information on cockroach bait aversion, review laboratory and field research, and offer recommendations for effective cockroach management stressing good IPM practices.

Influence of pest population structure and movement behavior on interpretation of pheromone monitoring programs in flour mills

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A long term monitoring project was conducted at a wheat flour mill. Results suggest pest populations interact over one of two spatial scales and this difference in scale is critical for effectively implementing IPM programs. For *Plodia interpunctella* and *Trogoderma variabile* at this location, pheromone trap captures outside were higher than inside, inside and outside captures were correlated, both indoor and outdoor trap captures cycled with season, fumigations did not reduce indoor trap captures, they were rarely if ever found in product samples, and marked *P. interpunctella* was demonstrated to enter buildings from outside. This pattern suggests that sources of insects lay over a spatial scale larger than the mill and movement effectively linking activity inside and outside the mill. *Tribolium castaneum*, in contrast, tended to have lower pheromone trap captures outside compared to inside, recovery in traps and product declined after fumigation and then increased until the next fumigation, inside trap captures did not follow a seasonal cycle, and was the primary species found infesting product. This suggests that source patches for this species lay over a spatial scale primarily contained within the mill itself with pheromone traps capturing primarily insects moving among these internal patches.

Response of insect grain pests to temperature choice tests

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Replacement of conventional chemical treatments with novel physical and biological methods to eradicate storage beetles requires better understanding of pest behaviour. Temperature is known to influence locomotory activity and population dynamics but its importance relative to other factors such as gravity and light is still not understood. Laboratory experiments are therefore being undertaken to establish how representative species respond to thermal gradients under various conditions. The movement of three species, *Cryptolestes ferrugineus* (Stephens), *Oryzaephilus surinamensis* (L.) and *Sitophilus granarius* (L.), was assessed under non-uniform temperature conditions. This was done first in 2-D arenas (390 mm long x 90 mm wide) where the only variable environmental cue was a temperature gradient (3 zones) generated in the floor of the arena. Later experiments used a 3-D arena where the 3 different zones were generated within sections of pipe (200 mm long x 70 mm diameter) filled with wheat at 60% rh. Results with the 2-D arenas have shown a hierarchy of attractancy for temperatures above 25°C as (in ascending order) *S. granarius, O. surinamensis* and *C. ferrugineus.* A wild strain *of O. surinamensis* showed a preference for low temperatures (15°C) but subsequent trials with the 3-D arena have reversed this result.

Some recent developments in wood preservation with respect to termite management

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Low-cost, general-purpose preservatives such as creosote, pentachlorophenol and water-borne inorganics have performed well, but are now under increasing scrutiny worldwide with regard to environmental, health and safety concerns. There is a global trend to replace such conventional preservatives and reduce the heavy metals (chromium and arsenic) which are contained in the most widely used water-borne preservative, copper-chromium-arsenic (CCA). Hence, the development and acceptance in some countries of alternatives such as ammoniacal copper quaternaries (ACQ's), copper azoles, CuHDO and other formulations. There is a trend to more specific niche-market preservative formulations, especially where safer and environmentally benign protection systems may be required. Boron compounds have made a remarkable reappearance, particularly for termite management, targeting either house framing or exterior timber products where resistance to leaching is required. The development of natural products to protect wood and wood products from termite attack is gaining momentum. Envelope treatments of wood with novel solvents and more environmentally-friendly actives that are functional under reduced penetration requirements are gaining acceptance. Protection of wood-based composites from termite attack by adding to the adhesive insecticidal additives of low mammalian toxicity is becoming more common. Interest continues in chemical modification and heat treatment of wood to impart resistance to termite attack.

OH BUGGER! THEY'RE BACK! The resurgence of bed bugs in Australia

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Over the last 50 years, with housing improvements and modern insecticides, bed bug infestations became a rare event. However, the situation has reversed in many countries, including Australia. Since 2001, we have recorded a 250% increase in bed bug samples. The Pest Control industry mirrors this trend, with one company reporting a 700% rise in treatments. The majority of the infestations has been in budget accommodation with high overseas guest numbers. There are likely many contributing factors for this resurgence. Globalisation has probably encouraged the spread of the insect via travellers and Australian Quarantine record the majority of interceptions from personal baggage via air travel from the Asia/Pacific region. We have recently identified the Tropical Bed Bug (*Cimex hemipterus*) that can invade warmer regions. Poor control operations have contributed to the problem. Our Department has been working with the Pest Control Industry and investigated a number of infestations. Treatment failure is a recurring theme, and many pest controllers have a poor understanding of the biology of this cryptic species and do not recognise and treat all harbourages. We foresee this trend will continue unless there is a concerted effort to inform the various stakeholders and develop more appropriate control methodologies.

Monitoring for parasitoids and stored-product moths

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In Germany and Austria, the control of the stored-product moths; Indian mealmoth *Plodia interpunctella* and the Mediterranean flour moth *Ephestia kuehniella* in food processing facilities and retail shops is achieved by releasing large quantities of *Trichogramma evanescens* Westwood using the inundative release strategy. These chalcid wasps lay their eggs into the moth eggs, thus killing the eggs and preventing the development of the pest. To assess efficacy, pheromone traps are used to monitor moth populations. To assess the effectiveness, and range of activity of *Trichogramma* spp., sentinel eggs are used. These are sterile moth eggs, placed throughout the facility, and collected at a later date and the parasitism noted. Monitoring the parasitoids with the help of sentinel eggs can also help to optimise the number and position of release points. The biocontrol of stored-product moths in Germany and Canada will be presented.

Use of acoustics in detection and studying activity of larvae of a soil nursery pest, *Otiorhynchus sulcatus* F. (Coleoptera: Curculionidae)

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The black vine weevil, *Otiorhynchus sulcatus* F., is a ubiquitous pest of ornamentals and small fruits throughout the Northern hemisphere, particularly, in areas where the environment is mediated by maritime and large inbound lake air flows N of 40° latitude. Primary damage to these crops is by the larvae that feed from late summer until the next spring on roots. Estimated damage in the state of Oregon is > $3 million USD/year. There are limited windows for control of these pests and a zero tolerance for plants that contain the pest. Detection for the past 150+ years has been by digging and sifting, limiting sampling quantity and efficiency. A portable hand-held acoustical detection device has been developed that can examine up to 15 container plants per hour. Pitfalls and successes and the feasibility of the use of acoustic detection for the European and North American nursery industry for this and other root feeding insects will be discussed. Information on the use of acoustics for activity studies of the larvae will also be discussed.

Acoustic detection of soil insects in pastures

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All too often, soil insect pests in pastures do significant damage before they are detected. Routine scouting techniques, which would involve excavation, would be too costly in this agro-ecosystem.. Modern acoustic technologies show promise as nondestructive tools for detecting hidden insects in the soil. Laboratory and controlled field studies, previously published, have shown acoustic detection methods can detect white grubs in the soil. A two-year study was designed to compare two estimates of soil insect infestation: acoustic detection and excavation. It was not expected that either method would give a perfect estimate of soil insect infestations. Soil insects tend to occur in patches. Destructive and non-destructive soil arthropods are often present in the same area. From a pest management point of view, identification of "hot spots" in the field are more important than perfect point to point correlation of acoustic and excavation data. The pros and cons of four data analysis methods will be compared: 1) point-to-point correlations of acoustic and excavation data, 2) point to point comparisons of kriged estimates of insect infestations (acoustic method or excavation method), 3) contingency table analysis, and 4) mapping of management zones, a precision agriculture approach, based on geostatistical analysis.

Long-term protection of Madam John's Legacy Museum in New Orleans, Louisiana, from subterranean termites using hexaflumuron

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The use of the SentriconÒ *Termite Colony Elimination* System for the control of subterranean termites in Madam John's Legacy House, a historic property in New Orleans, Louisiana, is described. A combination of in-ground (IG) and above-ground (AG) bait stations containing the active ingredient, hexaflumuron, was employed. A total of 208 g of bait matrix was consumed by the *R. flavipes* colony and it was eliminated by 21 October 1997. The stations became inaccessible in November 1997 due to a major renovation of the property. After the renovation, additional IG bait stations were placed in the soil through access holes cored through the concrete every 10 feet around the perimeter of the property and inside parts of the building with shared walls. Baiting resumed on 29 December 1998 with a total of 40 g of matrix consumed in the IG stations, and 229.2 g of matrix consumed in the AG stations. By 24 August 1999, only dead *C. formosanus* alates, soldiers, and workers were found in the AG stations and feeding stopped in the IG stations by 14 October 1999. This study demonstrated the importance of the AG component of the Sentricon System in structures that have little or no soil access and also illustrates the importance of controlling moisture-related problems in structures prior to the application of AG termite bait.

Insect pests in hospitals: Occurrence, possible risk they can pose for health and control methods

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Although in hospitals high level of pest management performance should be maintained, they are not pest free. The evaluation of insect pests occurrence in hospitals in Poland carried out in 2000-2003 showed, that 86,7% hospitals were infested by German cockroaches (*Blattella germanica* L.), 53,3% - by fleas, 20,0% by Oriental cockroaches (*Blatta orientalis*) and Pharaoh's ants (*Monomorium pharaonis* F.). Flying insects: wasps, mosquitoes, flies were present in 26,7%; 13,3% and 60,0% of examined objects respectively. Insects belonging to predominant species: *B. germanica*, caught in hospital wards and kitchens, were tested bacteriologically. About 80 bacterial strains were isolated from external parts of their bodies. Several of them, such as *Pseudomonas aeruginosa, Serratia marcescens, Citrobacter freundii, Enterobacter cloacae, Klebsiella oxytoca* are known as agents causing nosocomial infections. Susceptibility tests to selected antimicrobial agents pointed out, that several of isolated strains showed MLSb mechanisms of resistance (staphylococci), produced ESBL or/and Amp C-type beta-lactamases (Gram(-) rods). Such strains, resistant to antibiotics, may be reservoirs of resistance genes, which can be transmitted into other bacteria. Several isolated strains showed also resistance to surface disinfectants. The examined susceptibility to insecticides mostly used in hospitals showed considerable levels of insecticide resistance in tested strains of *B. germanica*.

Efficacy of *Cinnamomum* (Nees) oil and its major constituent against phosphine resistant and field strains of lesser grain borer *Rhizopertha dominica* (F.) (Coleoptera: Bostrichidae)

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The insecticidal activities of *Cinnamomum* oil and its major constituent zimtaldehyde against phosphine resistant and field strain adults of lesser grain borer *Rhizopertha dominica* (F.) were examined using feeding deterent and fumigation methods. Results show that both the components significantly ($p<0.001$) affect the mortality of adults of phosphine resistant and field strains. It also shows that phosphine resistant strain exhibits much more tolerant than the field strains for both the components indicating the higher lethal values. In a feeding deterent and fumigation test, zimtaldehyde was much more effective against the adults *R. dominica* than the *Cinnamomum* oil for both the strains. However, zimtaldehyde, major active constituent of *Cinnamomum* oil, may be suitable as fumigants of vapor-phase insecticides because of their high volatility, fumigations efficacy, and their safety.

Baiting systems for subterranean termite management

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Baits present a different paradigm for termite management. Rather than using barriers to exclude termites from a wooden structure, baits are designed to kill termites and reduce termite populations. Successful baiting should result in a building without termites threatening its structural integrity. Early baits incorporated sodium arsenite as the active ingredient. Refinements in the 1970s included use of decayed wood as an "attractant" and mirex, a slow-acting toxicant. Bait development accelerated in the 1990s; commercial products are now available. Many controversial assumptions and concepts underlie the baiting paradigm and include: random versus directed foraging, site fidelity, attraction, recruitment, and colony suppression versus elimination. Defining these ideas is demanding and has created spirited debate among researchers, chemical manufacturers, service providers, and regulators. Labor-intensive efforts, extended timeframe for the desired outcome, and conflict of interests in funding sources for research, also pose additional challenges to the large-scale adoption of subterranean termite baiting. The past and present history of baiting and current definitions of terms and underlying assumptions will be reviewed. Additional efficacy testing is needed using newer assessment technology, particularly the use of portable x-rays to view inside walls.

Paratransgenesis – a new approach to termite control

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Indigenous gut bacteria of the Formosan subterranean termite (*Coptotermes formosanus* Shiraki, Isoptera: Rhinotermitidae) were used as shuttles to deliver, express, and transfer foreign genes in termite colonies. *Enterobacter cloacae* was transformed with a recombinant plasmid containing genes encoding ampicillin resistance and the Green Fluorescent Protein (GFP). Termites from three colonies were fed with filter paper inoculated with transformed bacteria. Transformed bacteria were detected in termite guts by growing the entire gut flora under selective conditions and checking the cultures visually for fluorescence. We demonstrated that: (1) transformed bacteria were ingested rapidly and the GFP marker gene was expressed in the termite gut; (2) transformed bacteria established a persistent population in the termite gut; (3) transformed bacteria were efficiently transferred throughout a laboratory colony; (4) transformed *Ent. cloacae* were transferred into soil, however, they did not accumulate over time and the GFP plasmid was not transferred to other soil bacteria. Transgenic bacteria may be used to shuttle detrimental genes into termite colonies for improved pest control. To that end, we are screening gene products for their efficacy against termites and their obligate protozoan symbionts. Genes found to be detrimental will be engineered into natural gut bacteria and introduced to termites.

Contributions and possibilities of DNA biotechnology on termite management strategies at the dawn of the 21st century

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DNA biotechnology has had a major impact, particularly in the last six years, on the study of termite biology. An array of DNA marker technologies are being used to assess the temporal and spatial aspects of termite population structure, judge the efficacy of management strategies and unravel questions of taxonomy and gene flow. When used in collaborations with other scientific methodologies, DNA techniques have the potential to change the conceptual framework of termite management. Genomic and cDNA libraries have been developed and are being mined for DNA markers as well as developmental and structural genes that could lead to genetic-based management strategies that are environmentally and human friendly. Our presentation will, therefore, focus on: the historical development of DNA biotechnology for studying termite biology, the importance of that study for termite management strategies, and how DNA biotechnology may be integrated into termite management strategies for the 21st century.

High content of phenolic compounds in cocoa beans not utilised by storage pests

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Over the last decade, the consumption of cocoa (*Theobroma cacao* L.) has increased, however the yield of the crop in Ghana has been on the decline. To improve yield, alternative hybrids have been developed from crosses between Amazon, Trinitario and Amelonado genotypes to augment the Series II hybrids, which are already grown by farmers. Storage trials with these new hybrids have revealed that insect pest attack, which is very prevalent in dried / stored cocoa beans of the traditional type, is not a problem. Adults of *Tribolium castaneum* (Herbst) of the family Tenebrionidae and *Lasioderma serricorne* (F.) of the family Anobiidae, both Coleoptera, were introduced to dried cocoa beans and stored for 113 days at 33 ± 1°C and relative humidity of 72 ± 2 %. The beans of the hybrid varieties did not support the growth of the insects, which consequently died during the period of storage. In contrast, the traditional type supported the growth and multiplication of the insects and there was extensive damage to the beans. Analysis of the beans showed that the phenolic content of the hybrid varieties was higher than in the traditional type, and this may explain why there was an adverse effect on the insects.

Population genetics of Indian meal moth, *Plodia interpunctella*

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The Indian meal moth, *Plodia interpunctella* (Pyralidae: Lepidoptera) is a serious pest of stored food throughout the world. Its worldwide distribution is believed to have been accelerated by human commerce. It is also known to be resistant to a variety of commonly used insecticides. No information is available on the breeding structure of Indian meal moth at local, national, and global scales. Therefore, we developed polymorphic microsatellite loci to quantify the breeding structure and population genetics of Indian meal moth worldwide. Samples were obtained from selected counties within Kansas to investigate the fine-grained breeding structure. Samples were also obtained from several states in the USA and many countries in the world. Sampling is also being undertaken throughout the year at one storage facility in Kansas to understand the temporal variation in breeding structure. Results will be presented and discussed in the context of global movement of the Indian meal moth, implications for insecticide resistance management, and gene flow among subpopulations.

Perimeter treatment for subterranean termite control

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Pest Management Professionals had the opportunity to use newer, non-repellent active ingredients for subterranean termite control. These active ingredients provide control through reduction of termite populations. The pest control industry has asked if the newer non-repellents may provide acceptable subterranean termite control with less than full-conventional treatments. The study was undertaken to evaluate the applications with exterior perimeter plus localized interior treatments. In this study, 43 termite infested structures were treated with Termidor (fipronil) WG or SC using either an Exterior Perimeter Treatment (EP) or Exterior Perimeter with Localized Interior Treatment (EP/LIT). The 95% of structures exhibited no termite activity from 1st inspection date to 18-36 months after treatments. One EP site had recurring activity at 12 MAT at an interior area not treated at time of initial treatment. Another EP site had continuing activity at an interior site not treated at initial treatment through 6 MAT. All of the structures that received the EP/LIT which included treatments at the points of termite activity exhibited 100% control through the course of this study. According to these data, Pest Management Professionals can expect to use 19%-48% less diluted termiticide when using a EP/LIT approach versus a full conventional treatment.

Borates as a tool for termite management

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Borates are used worldwide as preservative treatments to wood and wood products. Formulations include aqueous solutions, pastes, gels, solid rods, powders, granules, glycol or paint mixtures, or mixtures with insecticides and fungicides or chemicals that enhance penetration into wood. Borates can provide resistance to damage by subterranean and dry-wood termites depending on their concentration and depth of penetration into wood. Their advantages are broad-spectrum toxicity to wood-destroying insects and some decay fungi coupled with low mammalian toxicity. Furthermore, borates typically are colorless, odorless, inexpensive, and practical to use. Because borates are water-diffusible, a major disadvantage is that they do not fix in wood; hence borate-treated wood is not suited for ground contact or exposure to weathering. For decades, Australia, New Zealand, and several European and South American countries have used borates to protect lumber, whereas the United States is a relative newcomer in this use. This presentation discusses laboratory and field trials to evaluate borate efficacy, as well as borate products and application methods. For example, in the United States an experimental borate bait demonstrated some success against field populations of a Sonoran Desert subterranean termite, and borate-containing plastic wraps around utility pole bases provided protection against termites and decay.

Termite baiting: a conservation and integrated termite management approach

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Termite control remains the biggest segment of business portfolio to the pest control industry. While conventional control methods such as residual spot treatment and chemical barrier are commonly practice, termite baiting technology since 1995 provides as an effective option to the industry and the public. Baiting technology is getting more popular as it controls/eliminates termite colony in less intrusive way to the property as compared with the post-construction method. In addition, significantly less chemical were used in treating termite infestation. Due to these, numerous invaluable historical sites infested with the subterranean termite were baited and restored in least damages manner. Most importantly, termite baiting plays a significant role in Integrated Termite Management (ITM) involving three continuous steps of monitoring-prevention-treatment, in addition to public education. While ITM concept has been long discussed and accepted in developed countries, it is now obviously changing the perception and practices of the termite control industry and the public of South East Asia and Greater China since introduction of baiting technology in this new millennium. The implementation challenges and solutions of ITM concept in these regions with specific references on hexaflumuron-based (Recruit®II) and noviflumuron-based (Recruit®III) termite baits are discussed.

Insecticide resistance in the German cockroach in Asia – problems and challenges

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Extensive usage and heavy reliance on the use of neurotoxic insecticides have led to the development of insecticide resistance in the German cockroach, *Blattella germanica* (L.), causing significant control failures to pest control operators world-wide. Despite being an important insect pest in Asia, reports on cases of insecticide resistance in field German cockroaches in this region had been limited to studies from Malaysia, Japan, Korea and Singapore. Most studies reported in the last 10 years are mainly from Malaysia. More than 80 field populations of this species that were collected between 1993 – 2000 showed broad spectrum resistance to all major groups of insecticides (organochlorine, organophosphate, carbamate and pyrethroid). Most of the collected strains possessed increased monooxygenase and esterase activities. Increased glutathione S-transferase activity was also found in a number of strains studied. The challenges of insecticide resistance in the German cockroach in Asia, and some of the management strategies adopted will be discussed.

Challenges to termite management with multi-genera faunas

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Termites are an important group of insect pests in the Asia Pacific region. Due to the high diversity of termite species in this region, it is common to find several termite pest species to co-exist, and to infest buildings and structures. In Malaysia, there are 12 species of subterranean termites from 7 genera (*Coptotermes*, *Macrotermes*, *Microtermes*, *Globitermes*, *Odontotermes*, *Schedorhinotermes* and *Microcerotermes*) that can be readily found in- and around buildings and structures, particularly in suburbia and rural settlements. Similar observations with species in the genera *Coptotermes*, *Microcerotermes*, *Macrotermes*, *Hypotermes* and *Odontotermes* are also recorded in urban and rural Thailand. Termites from three to six genera (*Mastotermes*, *Coptotermes*, *Schedorhinotermes*, *Heterotermes*, *Nasutitermes* and *Microcerotermes*) may be found within the same location as structural pests in Australia, with the highest number of genera in the tropical north of the country. Since the introduction of baiting in Malaysia, secondary pest species are more frequently encountered. Following elimination of the principal pest species (*Coptotermes* spp.) with bait, it is not uncommon to find species from other genera such as *Macrotermes* and *Schedorhinotermes* infesting the same building or structure after several months. Most of these species particularly those belonging to genera such as *Macrotermes*, *Globitermes* and *Odontotermes* from the higher termite (Termitidae), however, do not respond well to paper-based bait matrices. Options for managing multiple genera termite pest faunas in the tropics are discussed.

Documenting the novelty of non-repellent, liquid termiticide management strategies

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The advent of non-repellent termiticides in the 1990s was greeted with great scepticism in both the scientific and commercial market place, which had been preconditioned to thinking of termite control being exclusively predicated on repelling termite from structures. Since the coincident introduction of termite baiting systems and the launch of imidacloprid (Premise, Hachikusan), as a conventionally-applied liquid treatment, both the science and practice termite control have changed dramatically. This presentation will review the latest research documenting the pervasive effects non-repellent, liquid termite management strategies have on termites at both an individual and population level. Data on secondary kill in termite colonies, the mechanisms of transmission between termites, and the long-term impact on termite colonies/populations will be presented. The impact of these powerful performance factors on the protection of structures threatened with termite attack will be outlined. A discussion of what these new facts foretell about the future direction of termite management strategies will conclude the presentation.

Comparisons among technologies currently used for detection of hidden insect infestations in stored products

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Insect infestations are of major concern to food producers and consumers, but there are few reliable means of detecting insect pests hidden in packaged products before they cause significant damage. Modern acoustic and microwave radar technologies show promise as nondestructive tools for detecting hidden insects in stored products. However, it has been difficult to overcome problems resulting from background interference, reduced transmission of acoustic signals through multiple layers of packaging, and low levels of movement by some of the most important pests like Indian meal moth larvae. To meet these challenges, researchers and engineers have improved the sensitivity of detection probes and improved the interfaces between the probes and the stored products. They have also developed improved techniques for identifying insect signals in high-noise backgrounds. This report discusses some of the newest technology and potential advances that are in the process of development.

Bio-efficacy of gamma radiation and fumigants on *Tribolium castaneum* (Herbst)

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Integrated effect of gamma radiation and four commonly used fumigants (EDCT, ethylene dibromide, carbon disuphide and methyl bromide) was studied by exposing the larvae and adults of *Tribolium castaneum* (Herbst). Two regimens were tested, (i) irradiation followed by fumigation, (ii) fumigation followed by irradiation. It was observed that the larvae and adults exposed to 50 and 100Gy (nearer to LD_{50}) doses of gamma radiation, respectively, if fumigated within 6 hr after irradiation, required higher doses of fumigants at LC_{50} level, but when fumigation was performed 7 days after irradiation, relatively lower doses of fumigants were needed to achieve similar results. On the other hand, the fumigation of *T. castaneum* life stages followed by irradiation did not affect their radio-susceptibility. These studies indicated that if irradiation preceded fumigation, the pest susceptibility towards fumigants might be altered, wherein management of stored pest could be effectively operated. However, no evident change in the susceptibility pattern was noticed when the survivors of the fumigation treatment were exposed to gamma radiation. In view of these observations, the ionizing radiation might be considered as an environmentally sound alternative or supplement to other control methods.

Microsatellites and insect infestations: a new tool to investigate an old problem

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Microsatellites are considered the marker of choice for most population genetics studies because they are highly variable, codominant and reproducible. Microsatellites have been isolated and optimised for two psocid species, *Liposcelis decolor* and *L. entomophila* that are major pests of stored grain in Australia. Currently there is limited information on the invasion biology of *Liposcelis* species in Australia and an understanding of population structure and dynamics, gene flow among populations, geographic origin of source populations and dispersal mechanisms is required. Through the use of molecular methods a better understanding of the invasion ecology of *Liposcelis* species in Australia is possible and may assist in the development of strategies for the eradication, quarantine and potential future import of *Liposcelis* species or strains not present in Australia. To develop integrated control measures there is a need to understand the ecology of a pest in all its diversity. This will require a multidisciplinary approach utilising a diversity of methodologies. Molecular techniques such as microsatellites may provide a powerful addition tool to the field of stored product research.

Cost and efficacy comparison of IPM strategies with monthly spray insecticide applications for German cockroach (Dictyoptera: Blattellidae) control in public housing

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The long-term costs and efficacy of two treatments for German cockroach control were compared in public housing. The "traditional" treatment for cockroaches consisted of monthly baseboard and crack and crevice treatment (TBCC) using spray and dust insecticides. The integrated pest management treatment (IPM) involved vacuuming followed by monthly or quarterly applications of bait and insect growth regulators. Technician time and the amount of product applied were used to measure cost in both treatments. Trap catch (24h) was used as an indicator of treatment efficacy. The cost of the IPM treatment was significantly greater than the traditional treatment, particularly at the initiation of the test. However, after Month 4 the cost difference between the two treatments was no longer significant. To evaluate the costs of treatment over the entire year, technician time and product quantities were averaged over the total units treated. The average IPM cost was significantly greater than that of the TBCC treatment. While the TBCC was less expensive than IPM, it was also less effective. The TBCC treatment did not reduce cockroach populations. Trap catch in the IPM treatment was significantly reduced from 24.7 cockroaches per unit prior to treatment to 3.9 cockroaches per unit by Month 4.

Australian spiders: venomous villains or toxic treasures?

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"There are several groups of medically important araneomorph and mygalomorph spiders responsible for serious systemic envenomation in Australia. These include the red-back spider *Latrodectus hasselti* (Theridiidae) and funnel-web spiders from the genera *Atrax* and *Hadronyche* (Hexathelidae). The venom of these spiders contains potent neurotoxins that cause excessive neurotransmitter release. In the case of red-back spiders this is due to the action of a-latrotoxins to cause synaptic vesicle exocytosis, while d-atracotoxins from funnel-web spiders modulate voltage-gated sodium channels to cause increased firing of nerve impulses. In addition, cupboard (*Steatoda* spp.) and mouse (*Missulena* spp.) spiders appear to contain similar types of neurotoxins that may lead to systemic envenomation. Fortunately a number of antivenoms are available to treat envenomation from the bite of *Latrodectus* and *Atrax/Hadronyche* species, with extensive cross-reactivity within each genera and, in some cases, in unrelated families. In Australia, however, there is no evidence that spiders cause necrotic arachnidism despite many spiders being implicated including white-tailed (*Lampona* spp.), black house (*Badumna* spp.), and wolf (Lycosidae) spiders. Despite their notorious reputation, venoms of funnel-web spiders, have provided a untapped source of insect-selective neurotoxins (w- and k-atracotoxins) for the future development of biopesticides and the characterisation of previously unvalidated insecticide targets.
"

Advances in termite detection using acoustical technology

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An acoustical termite detection device was used to evaluate the presence of termites in the laboratory and field. Laboratory tests could detect differences in numbers of termites as well as detect changes in termite activity correlated with temperature differences. Field tests could detect termites in living trees. Differences in termite activity in trees were correlated with seasonal changes. Acoustical termite detection technology appears promising.

Tennessee and Kentucky stored grain project: an update on progress

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A cooperative effort with the University of Kentucky and the University of Tennessee was established in 2001 to determine if basic procedures and the use of existing pesticides labeled to control insects in wheat would be sufficient to prevent damage from stored grain insects in farm bins in each state. To this date there has been progress in reducing insect problems in demonstration grain bins using these established methods of insect control. This project is funded by an IPM grant to purchase necessary materials and monitoring equipment needed to complete the project.

New approaches to monitoring storage moths with semiochemicals

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Studies were conducted with Indianmeal moth, *Plodia interpunctella*, to help improve use of sex pheromone-baited traps for males and to elucidate the behavioral activity of a new female attractant. The role of pheromone quantity and quality on spatial targeting of an infestation source was studied in controlled experiments using a grid of regularly spaced traps and known numbers and release locations of male moths. Loading rates of synthetic pheromone on rubber septa from 1 to 2000 ug, or females producing natural pheromone, had no affect on overall re-capture rate of released males or on the ability of trap-captures to indicate the location of the release site. Other experiments investigated the role of trap height and position on trap catch and showed that male moths preferred to orient to a pheromone source associated with a physical surface, such as the floor, ceiling, wall or a landing platform. An attractant for female *P. interpunctella* was developed that consists of a mixture of naturally occurring compounds from attractive food sources used for oviposition. Experiments on biological and environmental factors affecting response to the female attractant, as well as discussion of the application of this attractant to monitoring and suppression, will be presented.

Control of eastern subterranean termite *Reticulitermes flavipes* (Kollar) in Hamburg, Germany

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Reports of the eastern subterranean termite *Reticulitermes flavipes* from Hamburg can be dated back to the 1930ties. Control efforts in the fifties and eighties of the last century involved massive application of biocides into building walls and the surrounding ground, but only with temporal and limited success. Modern subterranean termite control is implemented by baiting techniques, which are specifically targeted and through which the amount of applied pesticide is limited to a minimum. Laboratory tests have shown that the attractiveness of wooden baits can be significantly increased through a predecaying process of the wood by the brown root fungus *Antrodia vaillantii*. Field data taken at the Hamburg site have shown, that *R. flavipes* can be successfully lured out of their natural feeding areas to brown root treated wooden stakes. Once these are accepted by foraging workers they are replaced by termiticide pressure treated stakes. Pesticide application is 0.5 ppm or lower. These low concentrations of the pesticide act lethal after trophallaxis and accumulation over time in the target organisms of the colony, the primary or secondary reproductives. The termite population in Hamburg was reduced to residual 5 % of its original size during the past 4 years.

The granary weevil, *Sitophilus granrius* L. (Coleoptera: Curculionidae) – a coevolutionary event with the dawn of neolithic agriculture?

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Among all stored-product insects the granary weevil is the only species never recorded outside storage facilities. Anatomical, physiological and behavioral aspects of recent and hypothetical ancestral species in the genus *Sitophilus* are presented and discussed in terms of adaptation to anthropogenic storage of grain. Preadaptations like full development inside the host kernel, endosymbioses with bacteria, and the reduction in flight activity to prevent water loss in a dry environment could have resulted in the evolution of a full synanthropic grain pest of cosmopolitan distribution. Faunistic, archaeological and historical evidences of the pests origin and spread in conjunction with early agriculture is reviewed.

Can pheromones and hormones be effectively used for termite management?

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In termites, trail following chemicals are the most studied group of pheromones. Dodecatrienol and its variants appear to be the common theme in termite trail pheromones. Whereas one can envision their role in trail following among above ground foragers, it is not clear how such pheromones can be effective for the subterranean species that live and forage in well-established enclosed chambers and tunnels. In some species, both trail and sex pheromonal properties have been attributed to the same compound, perhaps at different concentrations. Among hormones, juvenile hormone (JH) and its effects have been studied in some detail. JH appears to be involved in egg maturation in reproductive forms as well as regulation of soldier formation. A possible use for pheromones and similar chemicals for termite management would be to increase the attraction of termite workers to bait stations. Sex pheromones, to attract and trap adults of one sex or confuse mate finding may be feasible in species producing relatively small swarms. JH and its analogs might be used to disrupt caste ratios leading to the death of a colony. Genes of peptide hormones, such as a diuretic hormone if discovered could be cloned into a suitable shuttle vector for delivery to a termite colony and disrupt the fragile water balance of these desiccation sensitive species.

Monitoring *Liposcelis* species (Psocoptera: Liposcelididae) in bulk grain stores

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Psocids, *Liposcelis* species (Psocoptera : Liposcelididae) have in the last decade become much more important as pest of bulk grain storage in Australia. To understand biology and behaviour and to assess efficacy of control measures effective means to monitor populations are needed. To monitor *Liposcelis* populations infesting open topped bins of wheat and barley, three monitoring methods were assessed – pitfall probe traps inserted into grain, cardboard crevice trap (made from piece of corrugated cardboard 100x150 mm) and counting psocids found on a marked out target area – typically a handrail above the cell top. Cardboard crevice traps placed on concrete surfaces above the open cell top were found to be more sensitive that the same trap placed on the grain surface or pitfall traps. Catch also agree closely with mean numbers of psocids seen on handrails nearby. Handrail counts need to be repeated many times to give analogous results as counts very considerably within very short time frames. Cardboard traps are very cheap, easy to deploy and very sensitive. They are now in widespread use by the Australian grain storage industry to detect and monitor populations of *Liposcelis* spp.

Investigations into the early detection of phylloxera infestation using remotely-sensed and field spectrometry

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Phylloxera (*Daktulosphaira vitifoliae* Fitch) is an aphid-like, root-feeding insect whose devastating impact on the grape and wine industry is well documented. Once a vineyard is known to be infested with phylloxera, the only long-term management strategies are to replant affected vines with resistant rootstock and to establish quarantine protocols. In Australia, there is considerable interest in mapping phylloxera infestation as early as possible so as to minimise the financial burden to vineyard owners/managers and to contain the pest to as small an area as possible. Grapevine canopies show symptoms of phylloxera infestation some 2-3 years after being infested; by this time the infestation most likely extends much further than observations (airborne or ground-based) might suggest. The key to the early, or pre-visual, detection of phylloxera infestation is an understanding of the spectral and chemical response of grapevines to the pest. This paper demonstrates the current capabilities of multispectral and hyperspectral sensors on airborne and satellite platforms to detect phylloxera infestation in vineyards. With the aim of identifying the spectro-chemical signature of phylloxera in grapevine canopies, detailed discussion is given on the results and analysis of field spectrometry and leaf tissue chemistry from field campaigns to vineyards in central and northeastern Victoria, Australia.

Mating disruption of two stored-product moth species: comparisons of population estimation

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A comparison of two separate indoor mating disruption (MD) trials, using two different methods for population estimations will be presented. Mating disruption trials were applied to two localities, one chocolate factory (major moth pest: the almond moth (*Ephestia cautella*)) and one flourmill (major moth pest: the Mediterranean flour moth (*E. kuehniella*)). Chem Tica, Costa Rica, kindly provided one-component MD-dispenser, which emit 1µg of Z9,E12-14: OAc per day. One MD-dispenser per 100m^3 was applied in the localities, changed every three months. The flour mill population was measured by counting trap catches, using pheromone traps and the chocolate factory population was measured by using water buckets with sticky inside lining. The almond moth has previously been reported as being attracted and caught in buckets with water. Complete trap shutdown was observed in the flourmill, however moths could still be seen flying and sitting on walls. The water traps, which do not compete with the MD pheromones in the chemical communication channel, showed declining catches after one month of MD trials, indicating that MD was effective in decreasing the mating frequency in the population. Problems in how to interpret trap catches with pheromone traps are discussed as well as the biological relevance of water for *E. cautella.*

Feeding response of subterranean termite, *Coptotermes curvignathus* Holmgren (Isoptera: Rhinotermitidae) to *Azadirachta excelsa* (Jack) Jacobs (Meliaceae) extractives and its timber

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Feeding response of subterranean termite, *Coptotermes curvignathus* Holmgren (Isoptera: Rhinotermitidae) to *Azadirachta excelsa* (Jack) Jacobs (Meliaceae) extractives and its timber was studied. The result shows that both methanol and hexane extracts from leaves and barks significantly inhibited feeding activity and survival rates of the termites. At 1000 ppm, the amount paper consumed was reduced by 27%. The mortality rates from leaf-hexane and leaf-methanol extractives were 26.6 and 17.1%, respectively, and from bark-hexane and bark-methanol were 34.4 and 28.9%, respectively. Mortality from the control was less than 10%. A choice study conducted in laboratory and field shows that timber from *A. excelsa* was the least preferred by the termites. Its susceptibility was equivalent to that of a medium hardwood, *Koompassia malaccensis* or three and four times less susceptible than that of *Hevea brasiliensis* and *Pinus caribaea*, respectively.

Food preference of the potato tuber moth and effect of three non-cultivated Iranian plants against female egg-laying on stored tubers

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Potato tuber moth, *Phthorimaea operculella* Zeller (Lepidoptera: Gelechiidae) was introduced to Iran in 1968 and has spread throughout the potato growing regions in many provinces including Guilan. Oviposition preferences on five cultivated host plants, potato, tobacco, tomato, pepper and egg-plant, were tested in the laboratory and in the field. Of the plants tested, tobacco was preffered host. Oviposition deterence of leaf powder and aqueous extracts of three non-cultivated plants, *Sambucus ebulus* (Caprifoliaceae), *Artemisia annua* (Asteraceae), and *Pterocarya fraxinifolia* (Juglandaceae) which are cheap, safe and easily available to the farmers, all inhibited egg-laying on stored potato tubers. The extract and leaf powder of *P. fraxinifolia* was most effective deterrent to females egg-laying on potato tubers (95 and 82%) followed by *A. annua* (94 and 54%) and *S. ebulus* (65 and 14% respectively)

Progress in post harvest and structural fumigation

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Times of change are times of progress. With the phase-out of methyl bromide, progress in the post harvest and structural fumigation markets is being achieved with an emphasis on Precision Fumigation^{TM}, defined as optimizing fumigant use to maximize efficiency and minimize risk. An enhanced level of professionalism is being achieved with new fumigation products, treatment strategies, equipment, training, and fumigator attitudes. In the last few years, new fumigant products have been introduced: Eco$_2$Fume® and Vaporph$_3$os® (Cytec, West Paterson, NJ) two gaseous phosphine products and ProFume* gas fumigant, sulfuryl fluoride (Dow AgroSciences LLC, Indianapolis, IN). Several other fumigants are in development, including carbonyl sulfide, methyl iodide, and cyanogen (ethanedinitrile), but regulatory hurdles and development timelines are not clear. Also, new continuous reading fumigant clearance devices are now available. The phaseout of methyl bromide has stimulated research in postharvest pest ecology in grain storage, flour mills, and food processing plants, thus improving the timing, efficiency and economy of pest management programs. Stewardship and training requirements for fumigants have been enhanced, either mandated by government agencies or required by manufacturers. Overall, the level of fumigator expertise is improving as they investigate alternatives to methyl bromide, modern fumigation tools and more efficient fumigation practices.

Biogas application to check infestation by pulse beetle, *Callosobruchus maculatus* (Fab.) in stored green gram, *Vigna radiata* Linn.

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Biogas (methane: carbondioxide = 60: 40), which is abundantly available at farm houses in Punjab state of India, was evaluated for its efficacy to check infestation by pulse beetle, *Callosobruchus maculatus* (Fab.) in stored green gram, *Vigna radiata* Linn. The gas was passed at flow rate of 8.8 litres/minute through the grains (Var. ML 613) stored in PVC bins each of 100 Kg capacity (70 x 48 cm); average volume and time taken to fill a bin with the gas were 39.6 litres and 4.5 minutes, respectively. Before passing the gas, known-age eggs, grubs, pupae and adults contained in plastic containers (10 x 5 cm), were placed at the bottom, middle and top layers of grains in the bins. Exposures periods of 2, 4, 6, 8 and 10 days were given. Though, all the adults were killed with an exposure period of only 2 days but the 6-days' exposure resulted in 100 % mortality of both the eggs and the adults; larval and pupal stages harbouring inside the grains, however, remained insensitive even to an exposure period of 10 days. Germination and cooking qualities of the fumigated grains at the 6-days' exposure did not show any adverse influence on the grains stored over a period of 3 months during the peak activity period of the beetle (July to September) including rainy season.

On-farm research trials on utilization of solar energy bed for control of pulse beetle, *Callosobruchus maculatus* (F.) infesting gram, *Cicer arietinum* (L.)

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On-Farm trials on utilization of solar energy bed for control of pulse beetle infesting gram were conducted in rural and urban houses. Earlier results on subsequent increase in infestation level of 10 ,20 and 30% due to pulse beetle in 2kg gram indicated that three exposures on solar bed each for 5,10 and 15 minutes at 60,120 and 180 days storage ,respectively, excluding control , control in increase in gram infestation level was comparatively much less than in unexposed .Results on subsequent control in increase from 30% gram infestation level by pulse beetle after three exposures on solar bed for 5 (45.2, 57.0 and 69.5%), 10 (38.0, 45.0 and 56.0%) and 15 minutes (31.5,32.1 and 31.9%) were better than control i. e .82.8 ,96.5 and 98.9% at 60,120 and 180 days after storage. Three exposures on solar bed for 15 minutes proved better than exposure of 5 and 10 minutes at 60,120 and 180 days after storage. 37 demonstrations on preparation technique of solar bed for adoption were given to women and men in rural and urban areas. Villagers took interest in solar bed preparation. Low cost technique of exposure on solar bed at regular intervals will eliminate beetle population and enhance quality of stored pulses .

Acoustic detection of Asian long horned beetle (Coleoptera: Cerambycidae) in wood: evaluation of factors affecting the spectral characteristics of vibrations resulting from larval feeding

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The use of acoustics to detect vibrations produced by insects living within concealed environments has been explored, including those living in wood. Insects feeding in wood produce vibrations by break wood fibers, and it are these vibrations that are of interests. However, a number of factors potentially affect the resonance properties of wood, including those inherent to the tree, the insect and the environment in which the tree is grown. Thus, studies were conducted to determine the effects of wood density, tissue, temperature and moisture, and larval instar on the frequency spectral characteristics of vibrations emanating from trees as they are being fed upon by the Asian Longhorned Beetle (ALB) larvae. This study represents an effort to capture the full range of the frequency spectra resulting from the breaking of wood fibers by ALB larva feeding within live host trees across the range of host tree species, tissues and environmental conditions in which ALB larva are found feeding under natural conditions in nature. As such, the goals of these studies were to develop a thorough characterization of ALB larval acoustical feeding signatures and to then use the resulting library to build a neural network.

Tunnelling geometry of subterranean termites and its implication in population management

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Understanding the food searching and acquisition behavior by subterranean termites is vital for managing their population, but it has been a challenging subject due to the cryptic behavior of these insects. Recent studies using laboratory foraging arenas indicated that tunneling geometry has a fractal dimension, and that termites were "attracted" to decayed woods and the extracts, possibly by the water soluble components of the decay wood extracts permeated in soil. The presence of sound woods, however, did not influence termite tunneling. Studies using a computer simulation program that was developed to identify critical components of tunneling geometry suggested vector navigation and path integration for subterranean termites. One evident benefit of these findings is the better placement of baits for the population control, but they may also aid the development of other control strategies.

Traps and product samples as indicators of insect activity in flour mills

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Types and numbers of insect species associated with flour mills were monitored at Kansas State University's (KSU) pilot flour mill and four commercial flour mills located in central Kansas and Nebraska. The pilot mill was monitored over a 29-week period from June to January 2002, while each commercial mill was surveyed for a two-week interval during July 7-31, 2003. Traps located in each mill were sampled every 2-16 days, using food- and pheromone-baited pitfall traps (Trécé) for beetles and pheromone-baited sticky traps for moths (85-192 traps/mill). In addition, pitfall and aerial sticky traps were also placed outside each mill (6-20 traps). Furthermore, product samples were collected from four categories; dirty incoming wheat, moving mill stock, processing equipment, rebolt sifter scalps, and spilled stock found on floors and other structural surfaces. Available samples varied by floor and week due to fluctuations in amounts available. Size of samples ranged from 0.2-1006.7 grams with sample sizes between 1-118 samples/floor/week. Results showed that the pilot mill found fewer species (9 families and 2 orders) while the commercial mills showed a more diverse population of insects surveyed (24 families and 5 orders). High levels of *Plodia interpunctella* (Hübner) and *Tribolium castaneum* (Herbst) were found in all five mills with *Tribolium confusum* (J. du Val), *Typhaea stercorea* (L.), and *Ahasverus advena* (Waltl) found at high levels only in the four commercial mills.
High numbers of fungus feeders suggest more ideal conditions for mold growth and a need for improved sanitation practices. In addition, it appears that fewer infested product samples were located in the commercial mills (7.11%) than the pilot mill (44.4%). It is believed that the commercial mills could either have lower insect densities, immigrating insect populations, or a sampling scheme that doesn't provide a complete representation of the total insect population.

A global perspective on nuisance ant control

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The world's population is becoming urbanized. In 1950, just 30% of the world's population lived in urban areas. By 2000 that percentage had swelled to 47%, and by 2030 the number is projected to be 60%. The United Nations predicts that almost all of the world's population growth by 2030 will be in urban areas. Urbanization often leads to problems directly associated with pests. Insects and rodents, for instance, infest and foul food at every stage. In this presentation, I will overview current trends in nuisance ant research and control globally. The most common methods of ant control include the use of toxic baits, liquid sprays, dusts, and granular formulations. Due to Central Place Foraging behavior exhibited by ants, control can be considered situational---that is, (1) direct nest treatment or physical removal when the nest is found; (2) baiting when only foragers can be found, and; (3) a combination of treatments when the nest cannot be found and foraging ants do not readily take bait or do not take enough to eliminate the reproductive capability of the colony. Finally, product formulation(s) and those area(s) that may need treatment are entirely dependent upon positive ant identification and thorough inspection resulting in an exact colony location or estimate thereof.

Detection of insect infestation in wheat transported in railroad hopper-cars

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Grain buyers typically decide whether to purchase a railcar of grain by taking a discharge sample from each compartment of a railcar and counting external adults or insect-damaged kernels (IDK) in the sample. To determine whether a discharge sample is representative of the level of insect infestation in a railcar, we examined levels of infestation in three-hopper railcars transporting wheat from country elevators to a mill. The most abundant species were *Rhyzopertha dominica* and *Cryptolestes ferrugineus* (84.5 and 14.2% of all insects collected, respectively). The immature stages of the two species were the most prevalent (> 95%). Number of insects or IDK in the discharge sample was not correlated with number of insects in the entire compartment. Numbers of immatures and IDK differed among railcars and compartments within railcars, but not among grain depths. The results indicate that each compartment of a railcar should be sampled to determine insect infestation, but that sampling at different depths within a compartment is less important; and that number of insects or IDK in the discharge sample is not representative of the insect infestation in the entire compartment. Grain buyers would benefit from development of a rapid technology for detecting internal insects in grain.

First report on the presence & medical importance of stinging ant in Southern Iran (Hym: Formicidae: Ponerinae)

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The importance of stinging ants, *Solenopsis spp*, in South and North America (Southern U.S) is well established. The report on the presence of stinging ants in Iran was quite astonishing to 50-years-history of Iranian Medical Entomology. The study was carried out since Nov. 2001 in Lar County in Fars and Sistan-Blouchestan Provinces. The specimens were collected from urban and rural habitats. The species was unanimously identified by colleagues at Natural History Museum of America in New York and Sam Huston State University in Texas, U.S., as *Pachycondyla sennaarensis* (Mayr, 1862). Local distribution of *P. sennaarensis* was surveyed in relation to urban and rural habitats where the majority of colonies were found in urban ranging between 1.24 colonies per 100 m^2 to 3 times more. A total of 152 persons out of 160 individuals who had experienced the sting of the ant, received medical treatment in local clinics and 6% of them were found to be highly sensitive. The stinging occurred indoor in 91% of the time in Lar city, in spite of the fact that 84% of the colonies were found in the yard and gardens and other places in houses.

Diffusivity (motility) of *Cryptolestes ferrugineus* adults in stored grain

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A method to calculate the diffusivity (motility) of the adult *Cryptolestes ferrugineus* (Stephens) (Coleoptera: Cucujidae) under a constant temperature or a temperature gradient was developed. The movement and distribution of the adults was governed by an advection-diffusion equation. To calculate the diffusivity of the adults in stored grain, published data were used.

Recent developments in the use of fungi for biological control of termites

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Biological control of termites has often been suggested as an environmentally safe control method. Subterranean termites live in an environment conducive to the growth of microbes and establishment of epizootics; however, few successful biological control agents have been reported. The Formosan subterranean termite (FST), *Coptotermes formosanus* Shiraki, frequently infests and causes large voids in living trees. A strain of the fungus *Paecilomyces fumosoroseus* (Pfr) was found to cause 100% mortality to FST and native subterranean termite workers when only a small percentage of the termites were directly exposed. Pfr can be efficiently produced as a wettable powder thereby affording several formulation and delivery options. Field application of a liquid suspension of Pfr to active galleries in infested trees resulted in reduction of FST activity within 12 days based upon video surveillance. Reduced FST activity was observed for an additional 157 days and observations are ongoing. An isolate of another entomopathogen, *Metarhizium anisopliae*, was recovered from dead alates. Purified spores of this isolate caused 100% mortality of FST alates at 3 days in laboratory tests. The most effective formulations of these pathogens, singly and in combination, for application to termites in trees are being investigated.

A non-chemical approach for the management of subterranean termites

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For many decades the principle method for preventing infestations by subterranean termites in residential and commercial structures has been with soil treatment termiticides. These pesticides ranged from the highly residual chlorinated hydrocarbons to the present third generation non-repellant compounds, and bait systems containing chitin synthesis inhibitors. However, as an alternative to chemical applications a number of physical barriers have been developed and tested for efficacy with several species of subterranean termites in the United States and Australia. This paper is a review of physical barriers that are currently commercially available in these countries.

Management of the Formosan subterranean termite: yesterday, today, tomorrow

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Subterranean termite management has been of interest for decades to researchers, private industry and homeowners since these pests have escalated in economic importance in urban environments. The increase in importance of these destructive pests evolved from the paucity of effective management tools. Early attempts were limited to inorganic pesticides as spot treatments for infestations and it was not until the 1940's that liquid treatment barriers were available with the residual chlorinated hydrocarbons. Soil treatments with this insecticide group predominated for more than forty years when other chemistries began to appear. Within the last two decades subterranean termite management has evolved to safer and environmentally acceptable soil termiticides, termite baits, and physical barriers. Good termite management also includes effective termite detection, development of which has transformed from the traditional probe and flashlight to sophisticated electronic devices that incorporate microwaves, the infrared spectrum, x-rays and acoustic emissions. It is foreseeable that future technology will lead to subterranean termite bait attractants, improved termite detection techniques, possible genetic population size determinations and gene manipulations that can modify termite behavior.

Seasonal & spatial changes in insect density in commercial elevator bins.

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Paper not available at time of production.

Adult survival and reproductive effects of forced egg-retention time in *Aedes albopictus* after application of deet as an oviposition deterrent

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The insect repellent deet (0.1% concentration) used as mosquito oviposition deterrent in the laboratory could influence the gravid female *Aedes albopictus* Skuse to retain and maintain mature eggs in her body. The egg-retention in female mosquitoes could benefit survival of the gravid females. The gravid females can lay the maintained eggs on availability of water or another liquid medium in the cage, but the length of forced egg-retention time could reduce the mean number of eggs laid per female. The gravid females that retained eggs also laid a higher percentage of non-tanning eggs, which increased with the egg-retention time. The rate (%) of egg hatch was not affected by deet used as oviposition deterrent. It was however, affected by egg-retention time in both treated (exposed to deet) and untreated (control) gravid females. The rate of egg hatch reduced after 3 weeks of retention, and after 6 weeks of retention the eggs of both treated and control gravid females could not hatch. The fecundity and fertility of gravid female *Ae. albopictus* was affected by the time duration of forced egg-retention.

Haematobia irritans **(Diptera: Muscidae) mitochondrial genome amplification and sequencing: potential of the long-PCR approach for calyptratae mitogenomics**

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Long-PCR strategies have significantly improved the sequencing and characterization of insect mitogenomes essentially by providing an alternative way to purify sufficient mtDNA from limited amounts of individual template and thus stimulating a wide range of otherwise limited techniques on insect molecular and evolutionary biology. We optimized Long-PCR reactions for the amplification and sequencing of the complete mitogenome of the horn-fly, *Haematobia irritans* (Diptera: Muscidae), a key parasitic fly of cattle. *H. irritans* mitogenome was amplified using four conserved Long-PCR primers designed on Dipteran 16S lrDNA gene combined with two previously described 'universal' primers. Amplified products ranged from 8.5 to 9.3Kb depending on primer combination, with a 1.5Kb overlap. We obtained specific products on a microgram range providing sufficient mtDNA for several techniques, including RFLP analysis and the sequencing of the *Haematobia irritans* mitogenome from a shot-gun library approach. Using H16S primers, we also amplified long-PCR products from seven other Calyptratae key species of medical-veterinary importance: the screw-worm fly, *Cochlyomyia hominivorax*, the blowflies *Chrysomya albiceps*, *Chrysomya megacephala*, *Chrysomya putoria*, *Lucilia cuprina*, the botfly, *Dermatobia hominis* and another muscid, *Musca domestica*. These results contribute not only to achieve complete *Haematobia irritans* mitogenome but also stimulate Calyptratae mitogenomics.

The complete mitochondrial genome of the human botfly *Dermatobia hominis* **(Diptera: Oestridae)**

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The botfly *Dermatobia hominis* is a parasite of humans and livestock, producing a furuncular myiasis. The larvae of *D. hominis* develops in the subcutaneous tissue causing economic loss and health problems, being one of the most important livestock pests in Latin America. The complete mitochondrial DNA (mtDNA) sequence of *D. hominis* was determined and has 16360 bp. The nucleotide composition is biased towards adenine and thymine, which accounted for 77.8% of the whole genome. The *cox1* gene has a Serine as the start codon, while incomplete termination codons were found for *cox1*, *cox2* and *nd5*. The control region, protein-coding genes, rRNA and tRNA genes are in the same order and orientation when compared to other dipteran mitochondrial genomes, except for a duplication of $tRNA^{Val}$ located approximately in the same position of the $tRNA^{Asp}$, but in the opposite strand. The identification of restriction sites on the sequenced mtDNA and the correlation with previous RFLP analyses provided insights for an overall scenario of the genetic variability and population structure of *D. hominis*, being helpful for monitoring the variation and dynamics of this species. The availability of complete mitochondrial sequences is an important source of sequence information for Diptera molecular and evolutionary approaches.

Are *Pediculus humanus* infected with *Wolbachia* spp.?

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Wolbachia spp. belong to the alpha-subdivision of the Protobacteria and are closely related to the Rickettsia and their relatives (Class Alphaproteobacteria, Order Rickettsiales). *Wolbachia* spp. are obligate intracellular bacteria and have been recorded in approximately 76% of insects studied. *Wolbachia* spp. are transmitted from mother to offspring through the cytoplasm of the egg. Horizontal and accidental transmission have also been recorded. The impact of a *Wolbachia* sp. on its host may vary. Enhanced fertility or fecundity, pathenogenicity, parthenogensis, feminisation, male-killing as well as cytoplasmic incapability (unidirection and/or bidirectional) may be caused by *Wolbachia* spp. infections. Cytoplasmic incapability has been linked to the transmission and inheritance of mitochondrial DNA in that it may provide the impetus for selective sweeps. The sweep of a mitochondrial-type through a population and from generation to generation may be enhanced in lice due to the short generation time, small population size and the founder effect that occurs as a result of dispersal of lice. Here we present our data on whether or not *Pediculus humanus* (body/cloths louse) and *Pediculus capitis* (head lice) may be infected with *Wolbachia* spp.

Triatoma mexicana in the state of Guanajuato, Mexico: preliminary results.

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Mexico have 7 genera and 24 species of Triatoma genus which are *Trypanosoma cruzi* vector's, some with habits intradomicile. In Guanajuato state have been reported 5 triatomines species, among them *Triatoma mexicana*, which is little studied. With the purpose of know their ecotopes, distribution, infection for *T. cruzi*, morfometric, citogenetic and of life cycle in the laboratory, 9 localities of three municipalities were studied in the northeast of the State. We collected 110 triatomines, adults 91(83%), nymphs 19 (17%), males 80 (73 %) and females 11 (10 %). Positive to *T. cruzi*. 5 (4.54%). Capture sites, 6 (4%) were captured indoors and 104 (96%) in the outdoors. The main ecotopes in the intradomicile were: bed, wall, kitchen light bulb and in the peridomicile were: external wall, courtyard light bulbs, stone wall and farmyard. This specie is attracted by the artificial light (tungsten). The season with more triatomines was spring. Studies of morfometric , citogenetic and biological cycle are not concluding. This vector doesn't present intradomiciliated habits like initially it had been reported, their biological cycle performance in the peridomicile is in the stone wall and farmyard in these sites we captured the biggest number of triatomines in the nymphs phase

Vertebrate integumentary chemicals: endogenously produced and acquired chemicals use to deter arthropods

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Ectoparasitic arthropods may be deterred from feeding on certain vertebrates by substances endogenously produced in the skin, pelage or plumage. Deterrent chemicals may also be acquired by them by rubbing against exogenous sources, i. e. anointing. Monkeys and birds rub themselves with millipedes that defensively discharge benzoquinones. Laboratory bioassays showed that three benzoquinones in millipede secretions deterred ticks and mosquitoes by repellency, toxicity and behavior alteration.

Spatio-temporal analysis of reinfestation by *Triatoma infestans* (Hemiptera: Reduviidae) following insecticide spraying in a rural community in northwestern Argentina.

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The spatio-temporal reinfestation patterns by *Triatoma infestans* following a blanket insecticide spraying in a rural community (Amamá) was analyzed using geographic information system (GIS), satellite imagery, and spatial statistics. Domestic and peridomestic reinfestation by triatomine bugs was monitored from 1993 to 1997 after the apparent elimination of *T. infestans* in 1992. In addition to domiciles, *T. infestans* occupied pig and goat corrals, wood piles, trees, and additional sites associated with chickens. *T. infestans* was detected at least once in 75% of 2110 sites evaluated. The prevalence of sites positive at least once for *T. infestans* during the study period increased sharply from 1993-1995 (0.6-2.9%) to November 1997 (32%). The initial source of *T. infestans* was a pig corral in southern Amamá one year post-spraying. Subsequent infestations were clustered around this initial focus at a distance of ~400 m starting in 1995. In 1996, clustering was maximized in sites within the same compound or in neighboring compounds at distances of 25-175 m. These results indicate that an effective control program on the community level will be based on the spraying of actual epicenters as well as sites within a 450 m of these epicenters to prevent the propagation of *T. infestans*.

Vector biology and control research in southern Africa

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In collaboration with the World Health Organization Regional Office for Africa in Harare, Zimbabwe, a multicentre study for the characterization of vector populations and capacity building for vector control in seasonal, epidemic-prone malaria transmission areas of southern Africa, was initiated. Namibia, Botswana, Zimbabwe, Swaziland and South Africa participated in mosquito collections, insecticide susceptibility testing and involvement in malaria transmission. *Anopheles arabiensis* was the major malaria vector in all five countries. Only Zimbabwe and South Africa gave indications of insecticide resistance in *An. arabiensis. Anopheles merus* was implicated in malaria transmission at one locality in central Zimbabwe. A further study on insecticide resistance in northern Kwazulu/Natal Province of South Africa was undertaken. The efficacy of spraying houses with both DDT and pyrethroids, either together as a single mix or one on top of the other was investigated as a possible resistance management tool. Capacity building was a major component of both these projects. Collaborators processed their own samples in the laboratory in Johannesburg, and two were registered for Masters degrees at the University of the Witwatersrand.

Identification of gene candidates for age determination in *Aedes aegypti* (Diptera: Culicidae)

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Aedes aegypti is the principal vector of dengue, a disease of increasing global significance. Vector population age structure is a critical determinant of a vector population's capacity to transmit disease. Current age-grading methods are of limited value as age estimates are restricted to 15 days, while only females older than approximately 14 days can potentially transmit dengue. Development of an age-grading method that predicts mosquito age over the entire lifespan of *Ae. aegypti* would be a valuable tool for disease transmission studies and evaluating vector control strategies. Here, we describe gene candidates that show age related expression profiles, for a rapid PCR-based method of age determination. Candidate genes have been identified from published *Drosophila* microarray data of gene expression across adult age. *Drosophila* candidate gene expression was confirmed using Northern blots of pooled RNA samples across a range of age groups (adults 1-29 d, with 4 d interval). Gene orthologues from *Ae. aegypti* were isolated by PCR using degenerate primers and gene expression profiles were validated by Northern blots analysis as for *Drosophila*. The application of real-time reverse transcriptase PCR techniques to determine gene expression levels in individual female mosquitoes and the statistical tools to predict age will be presented.

Detection and characterization of the tRNA^{Ile} in the mtDNA control region of myiasis-causing flies (Diptera: Calliphoridae).

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The family Calliphoridae is formed by myiasis-causing flies, being known for its economic, medical, sanitary and forensic importance. Ecological similarities and the lack of morphological diagnostic traits may cause problems in the identification of species, especially for larvae. The mitochondrial DNA (mtDNA) is providing important clues for species identification and variability characterization. The control region (CR) represents the major noncoding region of the mitochondrial genome, with a bias towards A+T content. The CR has been divided in two different subregions, including the A domain with conserved sequence blocks and a hipervariable B domain. The entire CR could be recovered in two separated PCR reactions using a specific and universal primers combination. The efficient PCR amplification, sequencing and analysis of nine Calliphoridae species has shown a complete tRNA^{Ile} duplication into the B domain of four *Chrysomya* species: *C. albiceps*, *C. bezziana*, *C. chloropyga* and *C. megacephala*. The tRNA^{Ile} duplication could be used as a diagnostic marker for *Chrysomya* genus identification, since the duplication has not been reported for other related Calliphoridae genus. The characterization of the A domain for Calliphoridae species will allow the evaluation of this region as a molecular marker for evolutionary studies and for identification of Calliphoridae species.

Parasitoids of nuisance flies on Australian cattle feedlots

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Populations of adult and immature nuisance flies and associated parasitoids were monitored on three Australian feedlots with one southern Queensland (SQ) feedlot being continuously monitored over two years. In the SQ feedlot both sentinel pupae (cultured *Musca domestica* exposed for 1-2 weeks at various sites) and feedlot pupae, consisting largely of *M.domestica* (85%) and *Stomoxys calcitrans* (12%), were examined for parasitoids. The 4.2% of the sentinel pupae parasitised produced 7 species of parasitoids, with *Muscidifurax raptor* (52%) and *Spalangia cameroni* (39%), predominating. Parasitoids emerged from 10.3% and 11% of the *M.domestica* and *S.calcitrans* feedlot pupae respectively, with the emergence pattern indicating that both species were equally parasitised. Unlike the parasitised sentinel pupae, for feedlot pupae *Spalangia* spp. constituted 90% of the parasitoids with *S.endius*, *S.nigroaenea* and *S.cameroni* comprising 50%, 22% and 18% respectively. *M.raptor*, *Trichomalopsis* sp and Diapriids made up the remaining 10%. Some parasitoid preferences for various feedlot sites were evident but *S.endius* was the most common species found at all sites. Parasitoids, largely *S.endius* and *S.cameroni* were seen in 9% and 12% of pupae from feedlots in central NSW and central Qld respectively. Parasitoids appear to be one of the more important biological control agents on Australian feedlots.

Successional aspects of the sarcosaprophagous arthropods fauna associated to rabbit corpses in the nearing areas to Tenjo (Cundinamarca)

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The succession of adult sarcosaprophagous arthropods was studied using baited Schoenly traps and rabbits as model. The efficiency of the traditional methodology (Ptrad) against the use of baited Schoenly traps (PTC) was comparated. Modifications to the Schoenly trap were made in the material of the trap, a communication with the substrate, interchangeable collector bottles and the collector funnels. During the 42 days of sampling 11.847 individuals were collected included in 99 species (97 in PTC and 63 in Ptrad); this includes 43 families (43 in PTC, and 32 Ptrad) belonging to five troffic rolls: necrophagous, opportunists, parasites, prayers and accidental. Also significant differences in the species abundance, richness, diversity and dominance were found between PTC and PTrad. By multivaried analysis techniques five stages were determined: Fresh (72 hours), Active decomposition (78h), Advanced Decomposition (330h), dry Remains (288h) and Bones (144h). These stages deferred in duration with those found in other works as a result of the sampling efficiency. Significant differences between the five stages were found in the abundance of individuals, species richness, diversity, dominance and community composition. In conclusion the PTC showed to be more effective in collecting arthropod fauna and superior to Ptrad for evaluating the adult sarcosaprophagous succession process.

Altered colonization of dung from cattle treated with doramectin, eprinomectin, ivermectin or moxidectin

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Endectocides (e.g., abamectin, doramectin, eprinomectin, ivermectin, moxidectin) are used worldwide to control internal and external parasites affecting cattle. After application, most of the dose is excreted in the faeces of the treated animal where residues can either enhance or reduce the attractiveness of dung to coprophilous insects. By influencing the number of colonizing insects, residues may indirectly alter the rate of dung pat degradation. The current study compared collections of insects from pitfall traps baited with dung of untreated cattle versus dung of cattle treated 1 wk earlier with topical applications of ivermectin, doramectin, eprinomectin or moxidectin. The effect of residues on colonization previously has not been reported for the latter three compounds. In addition, these results are the first to compare directly the effects of faecal residues on colonization, when potential confounding factors of drug formulation, cattle diet, insect species, and time are held constant.

On the ubiquity and phylogeny of *Wolbachia* in lice

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Wolbachia are intracellular bacteria of interest, because they profoundly affect the reproductive fitness of their insect hosts. Here, we provide the first report of *Wolbachia* in the two insect Orders Anoplura (sucking lice) and Mallophaga (chewing lice). We document the apparent ubiquity and diversity of *Wolbachia* in these orders with infections detected in each of 25 tested populations of lice, representing 19 species from 15 genera spanning eight taxonomic families. Phylogenetic analyses indicate a high diversity of *Wolbachia* in lice, as evidenced by the identification of 39 unique strains. Some of these strains are apparently unique to lice, whereas others are similar to strains infecting other insect taxa. *Wolbachia* are transmitted from infected females to their offspring via egg cytoplasm. Hence, the evolutionary radiation of *Wolbachia* strains is predicted to correspond with their host taxa. This prediction is met with strains in the *Wolbachia* taxon Supergroup B, which form a monophyletic clade. In contrast, strains in Supergroup A exhibit phylogenetic discrepancies that previously have been attributed to horizontal transmission between host species, possibly via a parasitoid vector. We interpret such discrepancies in the current study as evidence of genetic recombination between *Wolbachia* strains or between *Wolbachia* and non-*Wolbachia* bacteria.

Host preference in *Anopheles gambiae*: decision between plant and human volatiles in a wind tunnel

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Both sexes of *An. gambiae* have small energy reserves at emergence, and at 27^{o}C they begin to die within 2 days. Thus, 1 night after emergence is a critical time to obtain food. In a 2-choice wind-tunnel olfactometer, they oriented in significantly greater numbers toward honey, a source of nectar-related volatiles, than to human-foot volatiles. This was true both when the volatiles were paired with blanks and when they were presented simultaneously. In the latter case, the difference in response was greater, indicating direct competition between the two kinds of volatiles. After 5 days of sugar feeding and mating, males still strongly preferred honey volatiles, but females responded almost exclusively to human volatiles. These results suggest that under some conditions in the field, females take their first meal preferentially from plants. Females given sugar on the night of emergence exhibited stronger responses to human volatiles the following night, indicating that energy status itself promotes blood feeding, independent of maturation.

Phylogenetic relationships of midges of the *Culicoides obsoletus* species complex (Diptera: Ceratopogonidae) based on ITS2 rDNA sequences

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The midges of the genus *Culicoides* Latreille, 1809 include species that are vectors of orbiviral diseases of bluetongue and African horse sickness that cyclically decimate livestock in the Mediterranean Basin. Recent outbreaks of these diseases in regions where the known Afro-Asiatic vector, *C. imicola,* is absent has incriminated other *Culicoides* species belonging, amongst others, to the Obsoletus Complex of the subgenus *Avaritia* Fox, 1955. The lack of taxonomic clarity concerning these potential vectors stimulated the molecular analysis of 21 *Culicoides* population samples from 18 localities spread throughout mainland Italy. Nuclear ribosomal DNA ITS2 sequence variation was used to determine the genetic differentiation within and between the members of the Obsoletus Complex and related species, by means of AMOVA and phylogenetic analyses. Although morphological discrimination of the members of these complexes is extremely difficult, the molecular analysis clearly demonstrated a high degree of divergence between most of the species. Also evident is that the subgenus *Avaritia* comprises at least three species complexes, all of which are separate from the Imicola Complex which was represented by *C. imicola* as the outgroup. Furthermore, a simple PCR assay was developed which rapidly discriminates between the species of the complex, a prerequisite for vector identification.

"Differences in sensitivity to action of insect hormones analogues and agonists in several populations of *Culex pipiens* s.l. caused by insect pheromones"

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"Increasing interest for effective insect control, especially of hematophagous insects is provoked by number of diseases transmitted by some species from these insect groups. One of many ways how to control these species is the use of analogues and agonists of insect hormones. These analogues were tested against larvae of different strains of *Culex pipiens*. Larvae of *C. pipiens molestus* and *C. pipiens quinquefasciatus* originated from different countries were used in experiments. *Culex quinquefasciatus* is serious hygienic problem - vector of filariosis. Larvicidal activity of the methoprene, 20-hydroxyecdysone RH-5992 and RH-2484 were tested. All used compounds show different activity among experimental strains. In our laboratory strains - originated from Cuba, Iraq, India (two strains), Taiwan all tested compounds had dose depended activity. Toxic effect, morphological effects - inhibition of larval pupal or pupal-adult transformation and delayed pupation were observed. Very high activity of methoprene and 20-hydroxyecdysone were found in Cuban strain. On the other hand RH compounds were very active in Taiwanian strain. For example 20-hydroxyecdysone was still very toxic at 5 ppm concentration in Cuban strain, while in the Indian strain same toxic effect was caused at concentration 100 ppm. Similar results were obtained with methoprene RH-5992 and RH-2485.
"

Gene expression in ticks induced by blood meal and lyme borreliosis spirochetes

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The castor bean tick, *Ixodes ricinus,* is the principal European vector of *Borrelia burgdorferi* sensu lato, the causative agent of Lyme borreliosis (LB). Differentially expressed genes that underlie the host response of *I. ricinus* to *B. burgdorferi* have not been investigated prior to this study. The characterization of genes that are induced following an infected blood meal is essential for gaining an understanding of the molecular mechanisms that underlie vector-pathogen interactions. Subtractive hybridization was used to isolate tick genes that were induced in whole ticks following a blood meal and a blood meal on a *B. burgdorferi* infected guinea pig. Novel cDNA clones with similarity to cytochrome c oxidase, salivary secreted protein, actin, glutenin and a cysteine protease pro-peptide were induced following a blood meal. Novel cDNA clones with similarity to thioredoxin peroxidase(s), dolichyl-phosphate glucosyltransferase, glutathione-S-transferase, defensin, ML domain containing protein and von Willebrand factor were induced following *B. burgdorferi* infection. Northern analysis and RT-PCR were used to verify that these genes were differentially expressed in ticks and to determine the site of their expression. (Supported by the Grant Agency AS CR No. A6022306.)

Immune proteins in the soft tick *Ornithodoros moubata*

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Three proteins playing a role in the immune system and/or pathogen transmission were isolated from the soft tick *Ornithodoros moubata*.:1) An abundant tick plasma protein was isolated and identified as a protease inhibitor of a_2-macroglobulin (a_2M) family. Tick a_2M - **TAM** has an atypical structure in that it is composed of two noncovalently bound subunits of about 180 kDa. Each subunit is post-translationally processed in two disulfidically bound chains in a manner similar to the C3 and C4 components of complement. TAM sequence is closely related to the *Limulus* a_2M. TAM possesses at least 4 variants of bait region domain created likely by alternative splicing. 2) A sialic-acid binding lectin named **Dorin M,** apparently produced by tick hemocytes has been purified from tick plasma. Cloning and sequencing of the DorinM gene revealed that the protein shares significant similarity with the family of fibrinogen-related proteins (especially tachylectin 5A, B from the horseshoe crab and ficolins of vertebrates) which function as pattern-recognition molecules. 3) An anti-bacterial protein **lysozyme** was isolated from gut contents of engorged ticks. The enzyme is apparently upregulated upon bloodmeal. The features of lysozymes with anti-bacterial as well as digestive function seem to be combined in the tick gut lysozyme sequence.

Characterization of hemolipoproteins from ticks.

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Hemolymph proteins in ticks have not been completely studied. The hemolymph protein most studied in ticks has been vitellogenin (Vg). Besides Vg, another major hemolymph storage protein, a carrier protein (DvCP) and heme-binding lipoprotein (HeLp, for Heme LipoProtein) has recently been studied in two hard tick species but not in soft ticks. DvCP from *Dermacentor variabilis* is identical to a heme-lipoprotein (HeLp) from *Boophilus microplus*. An analogous protein to DvCP was reported in the soft tick, *Ornithodoros parkeri* (OpCP) but its tissue distribution and biochemical properties have not been fully understood. Further characterization of OpCP and biochemical characterization of major hemolipoproteins from other important tick species is in progress. Biochemical and molecular comparison of major hemolipoproteins across tick species and their significance to tick biology will be discussed.

Molecular detection of several tick-borne pathogens from ticks in Japan.

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Detection and analysis of several tick-borne pathogens, including *Rickettsia, Ehrlichia, Hemoplasma, Borrelia* and *Babesia,* from ticks recovered from dogs and cats in Japan were attempted using molecular techniques. A total of 1136 tick samples from dogs and 132 from cats were examined for tick-borne pathogens by using genus- and species-specific PCR, and sequence analysis. Some well-known pathogens such as *Anaplasma platys, Mycoplasma haemofelis, Mycoplasma haemominutum, Babesia gibsoni, Babesia canis* and *Borrelia garinii*, were detected and some new information on their distribution in Japan were added. DNA fragments of some new species were also found in the present study. Several rickettsial DNA closely related to *Rickettsia japonica, R. helvetica, R. canada,* or *R. akarii* were detected. Ehrlichial DNA closely related to a nevel *Ehrlichia* sp. detected in Tibet, Thailand and Africa were detected from *Hamaphysalis* ticks. Unique sequences that were similar to those of *Babesia odocoilei* and *Babesia divergens* were also obtained from *Ixodes ovatus*. *Borrelia valaisiana*-like DNA were detected from *Ixodes granulatus* in Okinawa. The combination of PCR and sequence analysis used in this study was demonstrated to be useful for detection of tick-borne pathogens from ticks. These can be excellent tools for epidemiological study of tick-borne diseases.

Species composition and dispersal of nuisance flies breeding on poultry farms in southern Australia

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"Ability to disperse and vectorial capacity makes flies a significant biosecurity and food safety risk on poultry farms. Although the species and biology of flies breeding on these farms has been widely studied overseas there have been few studies in Australia. We monitored numbers and species of flies breeding on egg farms in southern Australia over a 12 month period and assessed the dispersal of the major species using mass marking of resident fly populations with fluorescent dye. The little house fly, *Fannia cannicularis* and to lesser extent false stable fly *Muscina stabulans* were the major species for most of the year. In contrast to most overseas studies, *Musca domestica* was absent or present in only low numbers at most times. Most *F. cannicularis* remained within 700 m of the farm whereas marked *M. stabulans* were trapped at distances of up to 2 km. Modelling of total trap catches by distance predicted maximum dispersal distances of 1.6 km for *F. cannicularis* and 2.4 km for *M. stabulans.*

y-Season mosquito breeding associated with irrigation at Kununurra, Western Australia: potential impact on arboviral disease transmission.

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This study describes a preliminary investigation into the role of the Ord River Irrigation Area at Kununurra in mosquito production during the dry season. Specifically, the study aimed to test the hypothesis that irrigation has provided the opportunity for year-round breeding of arbovirus vector mosquitoes. A ten-day adult and larval mosquito sampling survey revealed that breeding was occurring during August, the iest month of the year at Kununurra, supporting the hypothesis that mosquitoes can breed year-round. Significantly larger numbers and percentages of adult *Culex annulirostris*, an important disease vector in Australia, were collected within the irrigation area (n=3706, 44.6%) compared to reference areas outside the irrigation area (n=363, 9.8%) (mean difference 76.2 per trap per night; 95% CI 38.6, 113.7; $p<0.001$). Larval *Cx. annulirostris* were also collected at several sites within the irrigation area, whereas none were collected at reference areas. Year-round mosquito breeding at Kununurra may increase human health risk from arboviral disease, as has been borne out by recent elevated notifications of Ross River virus disease in the late y-season compared to other towns in the Kimberley region. Mosquito management needs to be given a high priority to ensure this health risk is not further exacerbated. Strategies to reduce mosquito breeding should primarily focus on modification of breeding habitats and altering of irrigation protocols.

Comparative analysis of Calliphoridae complete mitochondrial genome sequences with other dipterans.

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In the view of the medical, veterinary, economic and forensic importance of Calliphoridae species, a comparative analysis of their nucleotide sequences with other dipterans would be useful for the molecular evolutionary studies of this family. Mitochondrial DNA (mtDNA) has been used mainly because of its maternal inheritance, accelerated rates of substitution, and lack of recombination. Of the 1020 Calliphoridae species described, 45 are listed in GenBank, including two complete mitochondrial genomes of the species *Cochliomyia hominivorax* and *Chrysomya chloropyga*. The comparison of the nucleotide content, skewness, aminoacid sequences, and codon usage between the mtDNA of Calliphoridae species and among the dipteran species can provide important features for studies in different taxonomic levels. Until now, the *cox1* gene is the most used molecular marker for studies among Calliphoridae and across a broad range of insects, but the comparison of individual and concatenated sequences of another mitochondrial genes among dipteran species have shown distinct degrees of conservation, indicating that some of them also should be employed as reliable molecular markers. The complete mitochondrial genomes are an useful source for improving primer designing, molecular identification of species, and for elucidate the potential of individual and combined genes for molecular systematic studies of Diptera.

Dispersal of house flies in the agricultural and urban environment: population dynamics Study using microsatellite markers

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The house fly, *Musca domestica* L., is a synanthropic species with worldwide distribution. Due to their unrestricted movement, mode of feeding, developmental habitats, and attraction to places where food is prepared and stored, house flies greatly amplify the risk of human exposure to food-borne and other pathogens. Flies that develop in animal productions where antibiotics are frequently used for prophylaxis and growth promotion carry antibiotic-resistant bacteria that maybe transmitted to food intended for human consumption, leading to illnesses that are difficult to treat. However, no studies have been undertaken to quantify the extent of house fly movement between rural to urban areas. Therefore, our objective is to examine the population genetics of house flies and estimate the extent of dispersal/migration from rural to urban areas and *vice versa* in Kansas. As a first step, we developed polymorphic microsatellite loci for house flies. Here we report on the isolation, characterization, and optimization of microsatellite loci and preliminary results on population dynamics of house flies.

Comparative study on the nocturnal behavior of *Aedes* mosquitoes using a new device equipped with photoelectric sensor

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The nocturnal behavior of non-blood fed *Aedes aegypti* and *Ae. albopictus* females was observed using a new automatic recording device equipped with photoelectric sensor. Carbon dioxide, heating and the contrast of black and white color were used for the attractive factors for mosquitoes. The nocturnal host seeking activity positively correlated to the increasing light intensity in both species. *Ae. aegypti* was found to be more sensitive to light than *Ae. albopictus.* The threshold of light intensity for activation of the nocturnal host seeking activity was < 0.1 lux (ca 0.01 foot candle) in *Ae. aegypti* and > 10 lux (ca 1 foot candle) in *Ae. albopictus,* respectively. Complete darkness in the daytime, on the other hand, deactivated the host seeking activity of *Ae. aegypti,* irrespective of their intrinsic circadian rhythm, which might well explain the importance of the sense of sight in *Ae. aegypti.*

Rainfall events determine the abundance of carrion feeding beetles in a West African savanna

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Using carrion beetles on goat and warthog skins as a model system, we studied the influence of single rainfall events on the abundance and flight activity of insects in a tropical savanna (Parc National de la Comoé, Côte d'Ivoire). Although the attraction of the skins declined with time (due to degradation and desiccation), the freshness of the resource was not the major factor determining the abundance of carrion beetles. After rainfall events the number of beetles sharply increased, even with older skins. We recorded the same effect in regularly watered skins (simulating continuing freshness), showing that the humidity of the resource was not responsible for the peaks. The sharp abundance peaks after rainfall events is of relevance for both ecological field studies which have often a standardized sampling programme ignoring weather, and for forensic studies relying on a predictable succession of carrion users which can be strongly influenced by weather.

Environmental parameters of habitat on breeding behavior of *Aedes albopictus* in Chonju, Korea

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Aedes albopictus during present studies has been associated with tree, automobile tires, very rarely apartment basements and open outdoor garbage containers. However, a contrast to the habitats of this insect is found; at one stage it seems that this mosquito can live in almost all places wherever there is debris, standing water regardless of the pH acidic or alkaline. Whereas, in certain cases it is evident that this insect is confined to a particular habitat, during the present investigation, it was quite evident that the outbreak of adult *Aedes* is very much dependent upon the percent humidity and temperature. The artificial habitats, that is abandoned tanks, cans, stray automobile tires if they are close to plantation or are in shade for most of the photoperiod they tend to be positive breeding sites for *Aedes* sp. In other situations, the same habitat in open with sunlight throughout the day tends to be negative breeding site. Water level in containers is also known to effect the population and species of the mosquitoes. Temperature and precipitation have been found to impose serious effects not only on the distribution of *Aedes* but also the size of the immature and adults as well.

Sheep lice control with entomopathogenic fungi

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Preliminary investigations into the potential for the biological control of the sheep body louse (*Bovicola ovis)* by the entomopathogenic fungi *Metarhizium anisopliae* and *Beauveria bassiana* are reported. Scanning electron microscopy was used to investigate the uptake of inoculum from the wool by lice and bioassay methodologies using a wool substrate were developed to test pathogenicity. Fungal strains were identified that caused a 100 % mortality of lice after 5days. The level of lanolin in the wool and stage of louse development had a significant effect on mortality rates but there was no effect of temperature within the range of 31^{o}C to 38^{o}C. Further studies are investigating factors that influence the pathogenesis of fungal disease in lice.

The mitochondrial DNA control region of Muscidae species (Diptera: Calyptratae): evolution and structural organisation

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The control region of insect mitochondrial DNA (mtDNA), also called A+T-rich region, is an interesting target for studies regarding the structure and evolution of the mtDNA. This non-coding region has an biased AT content, putative regulatory elements for transcription and replication processes, high levels of nucleotide substitutions and ins/dels events, and structural instabilities related to the organisation of internal sequence elements and its flanking genes. The sequence of *Haematobia irritans* and *Musca domestica* (Diptera: Muscidae) control regions have been determined and the sequencing of *Stomoxys calcitrans* control region is being conducted. Conserved sequence elements described for insect mtDNA control regions (such as poly-T streches, TA(A)n in tandem repeats, 'stem-and-loop' secondary structure and G+C-rich region) and conserved sequence blocks previously described for other Calyptratae species (myiasis-causing flies) have been identified in the Muscidae mtDNA control region. The share of such structural homologous elements among Muscidae and myiasis-causing flies suggests that Muscoidea and Oestroidea superfamilies have a similar regulatory mode for mtDNA transcription and replication processes. Intraspecific analysis of *H.irritans* populations using control region as a molecular marker are being conducted and length variation among specimens from Brazil, Argentina, Uruguay, Venezuela and Malaysia were already identified.

Key diagnostic characteristics of the third instar larvae of *Calliphora croceipalpis* and *Sarcophaga cruentata*

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Following the death of a human, various insects involved in the decomposition process invade the corpse. This invasion takes place in an orderly sequence, called insect succession. This poster deals with those morphological characteristics used to distinguish between third instar larvae of the secondary invader *Calliphora croceipalpus* (Jaennicke) and the tertiary invader *Sarcophaga cruentata* (Meigen). All aspects of larval morphology were studied, but specific note was taken of the following aspects for diagnostic purposes: nature of the spines and spine band, anterior spiracles, posterior spiracles and the spiracular atrium of the posterior spiracles. The distinguishing characteristics of *S. cruentata* were the deeply recessed atrium cavity and the scalloping of the inner margin of the posterior spiracular plate. In *C. croceipalpus* the distinguishing characteristics were the shallow posterior spiracular atrium and the orientation of the spiracles. These key characteristics were compared to the key characteristics of the third instar larvae of the other species that form part of this study *viz. Lucilia cuprina* (Wiedemann), *Chrysomya chloropyga* (Wiedemann), *Chrysomya marginalis* (Wiedemann) and *Chrysomya albiceps* (Wiedemann).

The effect of temperatue on the development of the immature stages of *Hemilucilia segmentaria* (Fabricius) and *Hemilucilia semidiaphana* (Rondani) (Diptera: Calliphoridae).

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Diptera larvae are among the most important carrion insects, because they are one of the most frequent and abundant organisms involved in animal decomposition process. This work objectived to determine the effect of temperature on the development of immatures of *Hemilucilia segmentaria* (Fabricius) and *Hemilucilia semidiaphana* (Rondani), two species of Neotropical blowflies commonly associated with animal carrion in rural and natural areas of Brazil, being two of the most important forensic indicators in the neotropics. The rearing of the immatures of both species was done using growth chambers with temperatures set at 10, 15, 20, 25, 30 and 35ºC, 12h photoperiod and 60 ±10% relative humidity. The mean developmental time from egg to pupation decreased as temperature increased. There were also differences in larval length, width and weight. These data were used to determine logistic growth curves to be used in estimating the age of the immatures of both species.

Larval competition differentially affects arbovirus infection and dissemination in *Aedes albopictus* and *Aedes aegypti*

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We determined the species-specific effects of intra- and interspecific larval competition between the mosquitoes *Aedes albopictus* and *A. aegypti* on adult susceptibility to infection with the arbovirus Sindbis. We found higher infection rates and whole-body viral titer, but lower dissemination rates, for *A. albopictus* than for *A. aegypti*. For *A. albopictus* but not *A. aegypti*, intra- and interspecific competition resulted in greater infection rates compared to low-competition rearing conditions. Size-dependent and size-independent effects of competition influenced arboviral infection and dissemination of *A. albopictus* in opposite ways. At a single level of competition (i.e., at a single density), higher viral titers occurred in larger *A. albopictus,* but between competitive treatments adults from low-competition larval rearing conditions had greater mean body size and lower body titer and a lower proportion infected, compared to adults emerging from high-competition conditions. Competition experienced by larvae had no detectable effect on acquisition of infection by *A. aegypti*. Higher dissemination rates and less sensitivity of *A. aegypti* females to effects of larval competition are consistent with the greater role of this species as a vector of viruses to humans.

Mitochondrial DNA variability in screwworm fly from Uruguay.

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The New World Screw-worm fly (NWS), *Cochliomyia hominivorax* (Diptera: Calliphoridae), is an obligate ectoparasite that causes primary myiasis in warm-blooded vertebrates throughout Neotropical regions. In Uruguay this pest is responsible for important economic looses to cattle breeding sector. In this work RFLP and PCR-RFLP analysis of mitochondrial DNA (mtDNA) were used to examine the genetic variability and population structure of six NWS geographic populations from Uruguay. RFLP analysis was performed using 5 restriction endonucleases and there were found 12 haplotypes. PCR-RFLP was used to survey mtDNA variation in control region and Cytochrome Oxidase subunits I and II. There were found 8 haplotypes utilizing 3 restriction endonucleases. The two techniques revealed the same pattern of haplotypes distribution, three haplotypes widely distributed and the remaining locally distributed. DNA diversity and divergence were estimated and the preliminary results revealed high genetic variability but no population subdivision. Our data are being compared with previous works that analyzed Libya samples collected during 1988 infestation in order to elucidate the origin of the introduction. The characterization of NWS intra specific genetic variability at the geographic level is necessary to monitoring the expansion in the current distribution and into screw-worm free areas. Financial support: FAPESP, CNPq, IAEA.

Paternal genome elimination in the human body louse, *Pediculus humanus* Linnaeus, 1758.

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Eight F_1 and two F_2 families of human body lice, *Pediculus humanus*, were bred to examine the inheritance of alleles in this species. Alleles at four autosomal microsatellite loci segregated in two ways in these lice: (i) in a Mendelian fashion i.e. diplo-diploidy, and (ii) in a non-Mendelian fashion. We interpret the latter as paternal genome elimination, where parental males transmit, exclusively, their maternal genome to their offspring (functional haplodipoidy). Further investigations of this extraordinary finding may provide insight into how haplodiploidy evolves in insects.

Anthropogenic ecological change and insects of health concern in New Zealand.

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Key components of anthropogenic ecological change include land conversion to human uses and the fragmentation and disturbance of native ecosystems. This is combined with the spread of alien species and human commensals and loss of locally evolved species. New Zealand is a microcosm of some global issues in this regard. Our research examines ecological change and changes in the habitat, distribution and productivity of native and exotic mosquitoes, the relation of mosquitoes and potential arbovirus vector species, and the disease burden of bee & wasp stings in New Zealand.

Paternal genome elimination in the human body louse, *Pediculus humanus* Linnaeus, 1758.

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Eight F_1 and two F_2 families of human body lice, *Pediculus humanus,* were bred to examine the inheritance of alleles in this species. Alleles at four autosomal microsatellite loci segregated in two ways in these lice: (i) in a Mendelian fashion i.e. diplo-diploidy, and (ii) in a non-Mendelian fashion. We interpret the latter as paternal genome elimination, where parental males transmit, exclusively, their maternal genome to their offspring (functional haplodipoidy). Further investigations of this extraordinary finding may provide insight into how haplodiploidy evolves in insects.

An *in vitro* larval immersion microassay for screening candidate Acaricides

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Acaricide resistance in *Boophilus microplus* is rising dramatically in regions of the world where this ectoparasite is prevalent, posing significant economic burdens on cattle production and productivity in Central and South America, South Africa and Australia. While *Boophilus* has been eradicated from the United States, acaricide resistance will increase the threat of reintroduction into southern portions of the U.S. New chemical agents are needed that combine potent activity with novel modes-of-action not exhibiting cross-resistance to existing acaricide classes. Currently, none of the available *in vitro* tick bioassays offers the flexibility, speed, and sensitivity required to rapidly screen large numbers of compounds for candidate acaricides. We have optimized and characterized a larval immersion microassay (LIM) that offers superior sensitivity, the flexibility to accommodate multiple solvents and detergents, and a robust capability for screening compounds in a relatively short period of time using a minimal quantity of test article for evaluation. Dose-mortality studies were conducted against multiple ixodid ticks, including ug-resistant *B. microplus*, with representative formamidine, organophosphate, synthetic pyrethroid, pyrazole, macrocyclic lactone and carbamate acaricides. The rationale for using this method to screen chemical or natural product libraries and identify, characterize, and prioritize compounds for further development will be highlighted.

Incidence of bed bugs (Hemiptera: Cimicidae) in shared Accommodation In Sydney, Australia

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Bed bugs have remerged as significant pests in Australia and worldwide in the past ten years, The results of a survey of the incidence of bed bugs in shared accommodation in Sydney is presented. The survey examined the incidence of the bed bugs and the relationship of the infestation to a number of factors including, season, building construction and furnishings. It also examined control methods, their frequency and effectiveness. The results demonstrated widespread incidence of bed bugs with the majority of premises reporting an infestation in the previous 12 months and with many premises having multiple infestations throughout that period. Current control measures did not seem to be eliminating the problem as recurrence of the pest was common. A more comprehensive control strategy for bed bugs needs to be developed to manage this pest in shared accommodation.

Characterization of the enzymatic activities present in extracts of various mites.

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Many of the previously-characterized allergens of house dust mites are known to be proteases and this enzymatic activity is thought to contribute to their allergenicity. Other astigmatid mites are also known to be allergenic but little or nothing is known about their enzymatic activities. The purpose of this study was to characterize the enzymatic activities present in extracts from 9 species of astigmatid mites, several of which have not previously been investigated. Extracts were prepared from 5 stored product mites (*Chortoglyphus arcuatus, Lepidoglyphus destructor, Blomia tropicalis, Tyrophagus putrescentiae, Acarus siro*), 3 house dust mites (*Dermatophagoides farinae, D. pteronyssinus, Euroglyphus maynei*) and one parasitic mite (*Sarcoptes scabiei*). ApiZym strips were used to screen for the presence of 19 individual enzyme activities. Digestion of 9 other substrates was also evaluated by spectrophotometric or electrophoretic methods. All mites exhibited some form of phosphatase, esterase, aminopeptidase and glycosidase activity although their substrate specificities varied considerably. Scabies mites did not possess detectable serine peptidase activity nor were they able to hydrolyze gelatin or casein while all other mites exhibited these activities. Storage mites possessed enzymes capable of degrading the widest range of substrates while scabies mites had the most limited proteolytic capacity.

Klinotactic orientation of the northern fowl mite to heat

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The northern fowl mite (NFM) (*Ornithonyssus sylviarum*) is a cosmopolitan bird ectoparasite and a serious pest of the U.S. poultry industry. Mite infestations spread within a flock, even when direct bird-to-bird contact is prevented, and heat is a suspected orientation cue. NFM movement patterns were evaluated in a spatial heat gradient, as well as in an environment with temporal fluctuation of temperature (spatially uniform). Mites were filmed and the movement tracks were digitally analyzed for changes in velocity, angular shift and mean bearing. NFM significantly increased linear velocity when exposed to heat (35 °C) relative to control mites in ambient conditions (~27 °C). Mean track bearings were significantly associated with the position of the heat source in a spatial gradient, while track bearings of mites in ambient conditions were randomly directed. Temporal shifts in temperature from heated to ambient condition caused NFM to execute turns ≥ 90° within 5 seconds. Shifts in temperature from ambient to heated condition did not consistently cause NFM to execute turns ≥ 90°. These experiments indicate NFM orient to a heat source by comparing temperatures at different points in time (longitudinal klinotaxis) and executing turns based on internally stored (idiothetic) proprioceptive information.

Screening medicinal plant extracts for effects on *Aedes aegypti* (Diptera : Culicidae) and toxicity to a non target organism

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A preliminary study was conducted to find out the effects of the extracts of 112 medicinal plant species, collected from the southern part of Thailand, on *Aedes aegypti*. Studies on larvicidal properties of plant extracts against the third and fourth instar larvae of *Ae. aegypti* showed fourteen species with high toxicity. Six plant species were comparatively more effective against the larvae at very low concentration. These extracts demonstrated no or very low toxicity to guppy fish (*Poecilia reticulata*). Three plants with promising larvicidal activity, *Mammea siamensis, Anethum graveolens* and *Annona muricata,* were used to study of the impact of the extracts on the life cycle of *Ae. aegypti*. The results showed that these plants affected the reproductive potential of surviving adults by reducing the number of eggs laid and egg hatchability. The first instar larvae were very susceptible to *A. muricata* and the second instar larvae were susceptible to *A. graveolens* while the third and fourth instar larvae were susceptible to *M. siamensis.* These extracts delayed larval development and inhibited adult emergence.

The prevalence rate of *Leishmania* infection among *Phlebotomus* collected from Abardejh of Varamin Iran

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Cutaneous Leishmaniasis is a prevalent tropical parasitic disease, which is caused by Leishmania protozoa. The infection can be limited in immun-compitant person, but it could progress to a chronic and ulcerative disease in immuno-comprmised person. The reservoirs are carnivore and rodents and the vectors are Phlebotomus. Abardejh of Varamin is located in southeast of Tehran and there is a tropical ecosystem in its eastern border. There is a suitable condition by the Tamarisk and rodents exist for *Phlebotomus* activity. In this study we investigated the prevalence of Phlebotomus population in Abardejh district. The collected sandflies by funnel trap from rodents' barrows transferred to the department of Parasitology at Pasteur Institute of Iran .The sampling was continued during the spring and summer in periodicals dates. The collected sand flies were studied to define the genius and species before parasitological culture in NNN media. The data revealed that the maximum activities of Phlebotomus were in Jun and July months. The results indicated the high prevalence rate of infection among the sand flies. This could be a matter of debate because of human and agricultural environments are located closely to this district. Therefore application of chemical control, environmental sanitation seems to be required to control of tranfection from sand flies to man.

The molecular identification of closely related calliphorids (Diptera: Calliphoridae) of forensic importance.

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Blowfly larvae (Diptera: Calliphoridae) are one of the most useful groups of insects utilised during the investigation of human deaths because they are among the first colonisers of a cadaver. The identification of such larvae is a critical step when estimating the the postmortem interval (PMI), as the rate of larval development can vary substantially between species. Two blowflies that have the potential to feature in forensic investigations are *Chrysomya semimetallica* and *Ch. latifrons.* The identification of these species is hampered by their almost indistinguishable morphologies, even as adults. The species were differentiated on the basis of two highly variable regions of the ribosomal DNA genes – the internal transcribed spacer (ITS) sequences. The amplification of ITS2 from both species was readily accomplished, however only ITS1 from *Ch. semimetallica* amplified successfully. The ITS2 differences correspond to 1.22 % sequence divergence between the species. A rapid, cost-effective strategy for species identification would involve PCR amplification of the ITS regions, followed by restriction fragment length polymorphism (RFLP) analysis. Despite the differences within ITS2, no restriction sites were observed that could differentiate the two species. Assessment of this possibility in ITS1 will be achievable once the remaining data is obtained from *Ch. Latifrons*.

A global presence of *Wolbachia* in human lice?

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Wolbachia are intracellular bacteria of general interest. The profound effects on the reproductive fitness of their insect hosts have resulted in plethora of phenotypes, which still await a full characterisation. Infections of *Wolbachia* in lice affecting humans are of particular interest. Endosymbionts of lice have a great potential for the control of these insects. Previous research in Canada suggests that infections of *Wolbachia* occur in all species of lice (Anoplura, Mallophaga). Independent and concurrent research in Wales provides substantiating evidence. The current study represents the subsequent joint efforts to test for the ubiquity of *Wolbachia* infections in populations of human lice (i.e, head lice - *Pediculus humanus capitis*, body lice - *Pediculus humanus humanus*, pubic lice - *Pthirus pubis*) from different continents.

Field efficacy of a personal insect repellent based on essential oils from Australian native plants against the mosquito (Diptera: Culicidae) and the biting midge (Diptera: Ceratopogonidae)

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Field trials were carried out in a wet tropical climate at separate sites in Cairns, Far North Queensland, Australia in Autumn 2002. One site consisted of a fresh water creek infested by mosquitoes and the other site a mangrove wetland infested by biting midges. Both studies were carried out during daylight hours and were conducted by periodically exposing human volunteers with bare lower legs (knee to ankle) to mosquitoes or biting midges over a four-hour period. Counts were made of mosquito landings over a five-minute period and biting midge landings over a three-minute period. Both legs of each volunteer were used to give five replicates of the repellent formulation and five replicates of the untreated control. Against mosquitoes, primarily *Verrallina carmenti* and biting midges, primarily *Culicoides ornatus*, the formulation significantly reduced the landing rate compared to untreated legs over the entire four-hour study period. This study demonstrated that this formulation provided protection against mosquitoes and biting midges for up to four hours after application.

Natural promastigote infection of *Phlebotomus caucasicus* in a new focus of visceral leishmaniasis in north west of Iran

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An investigation was carried out in Ahar County (a new focus of Visceral Lieshmaniasis) in order to determine the vector (s) of disease. This study was done in eastern Azarbaijan, North-west of Iran during the summer of 2003. Sand flies were collected using sticky papers, CDC light traps as well as aspirators. All of the collected sand flies were dissected in a op of normal saline for surveying of promastigote. Head and the two last abdomen segments of specimens were mounted in a op of Puri,s medium for species identification. Totally, 215 sandflies were collected including: *Phlebotomus kandelakii* (57.2%), *P.perfiliewi* (18.1%),*P.caucasicus* (11.2%), *P.sergenti* (7.9%), *P.papatasi* (3.3%) and *P(.adlerius)sp.*(2.3%). Only 1 out of 24 *P.caucasicus*(4.2%) was found naturally infected with parasites. This is the first infection report of *P.caucasicus* in this region. The ecology and some behavioural aspects of the vector will be discussed in details.

Application of a new *Aedes* surveillance method in Botucatu, Brazil.

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In Brazil, infection by dengue virus has increased significantly in the last two decades, after the reintroduction of mosquito vector *Aedes aegypti*. Epidemiological surveillance remains the only way to prevent dengue outbreaks. Currently, the control program applied in *Aedes albopictus* and *Aedes aegypti* populations are larval surveys. An interesting alternative is the using of a new control strategy named PCI (Premise Condition Index). This model relates the property condition, like house condition, yard condition, and degree of shade; to the occurrence of *Aedes sp* oviposition. The present study is based on the application of PCI in properties located in the urban area of Botucatu. Ovitramps have been set in 105 spots all over the city and their properties qualified. Results from properties analysis by PCI showed that 64.7% ones with scores 8 and 9 (considered bad condition houses) were positive to *Aedes albopictus* while only 19% of properties with scores 3 and 4 (good condition houses) were positive. This analysis demonstrated the accuracy of PCI method, since major occurrence of *Aedes sp* has been observed in properties with highest scores. There is a positive correlation ($r=0.76$, $p<0.025$) between PCI and positive house percentage, showing the significance of the method.

Molecular population genetics of *Aedes aegypti* in Brazil using RAPD

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Random amplified polymorphic DNA (RAPD) analysis technique has been used in studies of arthropod molecular taxonomy and in epidemiological studies of relatedness of geographic populations and vector movement. In Brazil, little is known about genetic structure among *Aedes aegypti* populations. We use RAPD to examine patterns of *Aedes aegypti* genetic structure in 11 populations from 6 Brazilian states. We found high genetic polymorphism (H_S = 0.38) and high levels of genetic differentiation exist both between population from different states (G_{ST} = 0.43) and between populations from cities in same state (G_{ST} = 0.41). These results indicate significant differentiation between *Aedes aegypti* populations in Brazil. There is a clear correlation between genetic structure and geographic localization showing that, if new populations are been introduced, they are not significant. Principal Component Analysis (PCA) of receive allele frequencies could separated populations in 3 different groups regarding geographic origin.

Biological characteristics of three domiciliated vectors of Chagas disease in Mexico.

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Mexico has a great population of triatomine bugs actually are reported seven genera and 24 species of Triatoma genus. Due to this great amount and diversity, surveys were made for to know ecotopes and biology of three species of the Triatoma genus. The objective is to show laboratory and fields observations made in regions where Trypanosoma cruzi vectors predominate: Triatoma barberi recollected in Queretaro state, T. pallidipennis in Morelos and T. dimidiata in Veracruz. T. barberi is an exclusive species of Mexico, it prefers walls, T. dimidiata is found in floor and the middle of the wall downwards, both in beooms, and T. pallidipennis prefers floor and clothes, also wildlife. Life cycle is 523 and 510 for Triatoma barberi and T. dimidiata, respectively and for to T. pallidipennis is 171 days. T. barberi feeds and defecates at the same time, the others two species defecated among 10 and 30 minutes. Infection index percentage was is 56.6 for T. barberi, 29.0 for T. pallidipennis and 15.7 for T. dimidiata. Percentage of infective forms in bugs feces were 76, 26 and 15 for T. barberi, T. pallidipennis and T. dimidiata respectively. T. barberi is the best vector of T. cruzi in Mexico

Droneflies occurring from effluent of manure in Hokkaido, Japan (Diptera: Syrphidae)

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Three genera and 5 species of droneflies (Diptera: Syrphidae), namely *Eristalis (Eoseristalis) cerealis, Eristalis (Eoseristalis) roccica, Eristalis (Eristalis) tenax, Eristalinus (Lathyrophthalmus) tarsalis* and *Helophilus (Helophilus) virgatus*, occurred from the ovitraps using the effluent of manure as attractant and media in Hokkaido, Japan. These droneflies were reared in the laboratory and the specimens of each developing stages were examined their morphological characters by the binocular and scanning electron microscopes. The following differences between the species were observed:

Bioconversion with *Hermetia* to solve problems of manure accumulations and produce valuable feedstuff

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Black soldier fly, *Hermetia illucens* can eliminate many of the problems associated with large manure accumulations and with other putrescent wastes. The larvae consume fresh waste rapidly in managed systems and assimilate nutrients that otherwise might produce odors, pest flies and cause environmental pollution. Mature prepupae are self-collected for use as an animal feedstuff with about 42% protein and 35% fat. As a ration component, prepupae meal supported better fish growth than otherwise similar rations using fish meal. Data will be presented on this and other feeding studies with prepupae meal, amino acid and fatty acid profiles, elimination or reduction of several malodorous gasses, control of pest flies and reduction of waste dry matter by 50% or more and reduction in concentration of potentially polluting nutrients within that residue.

Prevalence of dengue virus in *Aedes* mosquitoes during y season by semi-nested reverse transcriptase-polymerase chain reaction (Semi-nested RT-PCR)

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Dengue hemorrhagic fever remains a major health concern in Thailand. Much effort has focused on the prevention and control of the disease. Detection of dengue virus infection rate in mosquito would evaluate dengue control programs and predict the epidemics of dengue hemorrhagic fever. To determine dengue virus infection rate in mosquito by Semi-nested RT-PCR. A total of 400 mosquitoes were collected from Rom Kao Community representing a crowded community and another 9 non-crowded communities in Bangkok. Mosquitoes were then divided into 40 pools, each contained 10 mosquitoes. A total of 391 *Aedes aegypti* and 9 *Aedes albopictus* were screened for dengue virus. Mosquito infection rate in Rom Klao community is 2.5% of mosquito pool equal to that found in non-crowded communities. Both groups are found to be dengue virus serotype 3. Our study suggests a circulation of dengue virus serotype 3 in both crowded and non-crowed communities, the infection rates of which are indifferent during dry season.

Field evaluation of bifenthrin 10WP and bifenthrin 80g/L SC on dengue vectors at high-rise flats in Kuala Lumpur , Malaysia.

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Bifenthrin 80g/L SC and Bifenthrin 10WP were evaluated against the sentinel sugar fed adults and 4th instar larvae of *Aedes aegypti* at 17 storey high-rise flats in Kuala Lumpur.The impact of both pyrethroid formulations on field populations of *Aedes aegypti* and *Aedes albopictus* larvae was monitored weekly using bottle containers.Both formulations of Bifenthrin showed adulticidal effects causing between 96% and 98% of *Aedes aegypti* mortality in the field compared to 18-19% in the control. However,both Bifenthrin formulations had not shown larvicidal effects when sprayed with ULV. This field trial indicated that both Bifenthrin formulations had potential for use in future dengue vector control programs.

Geographical range predictions of *Anopheles* mosquitoes in northern Australia using a genetic algorithm and data mining methods

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An appreciation of the environmental factors which influence the geographical ranges of mosquito vectors of malaria is of considerable interest from both ecological and epidemiological viewpoints. GARP (the Genetic Algorithm for Rule-Set Prediction) was used for ecological niche modelling of *Anopheles farauti* ss, *Anopheles farauti* 2 and *Anopheles farauti* 3 in northern Australia. Models were developed using 41 environmental layers together with species locality datasets derived from systematic mosquito surveys in northern Queensland and the Northern Territory. For each species the predicted ranges included almost all presence records and excluded areas in which that species was not found in repeated collections. The datasets were analysed with CART (Classification and Regression Tree Analysis) and KnowledgeSeeker to investigate associations between environmental variables and the record/no-record collection sites for each species. The outputs were used for further model development with GARP to produce accurate range maps of *A. farauti ss* and *A. farauti* 3 using only 6 or 7 of the most significant environmental layers identified by decision-tree software. Further data mining analyses showed that the use of simulated absence data in lieu of no-record collection data may highlight significant environmental variables in datasets which do not have records for species absence.

A new pediculicide based on isopropyl myristate

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New pediculicides need to be effective, safe, and to act differently from other products so as to prevent or delay resistance. Isopropyl myristate (IPM) is approved for many cosmetic uses and is thus safe for topical use. IPM is not known to be neurotoxic to insects, unlike most other pediculicides. IPM in cyclomethicone was tested against body lice and their eggs (*Pediculus h. humanus*) as surrogates for head lice (*P. h. capitis*) by the ASTM methods. IPM gave 100% knockdown at +1 hour and 88.8% at +4 hours. 24-hour mortalities were: 15% IPM; 68.8%, 50% IPM; 97.1%, 100% IPM; 100%. Ovicidal action was slight, with mortalities under 20% The mode of action is unresolved, but is likely to be related to IPM's spreading and penetrating action. Symptoms were, however, similar to those from a neurotoxic synergized-pyrethrin pediculicide; Rid®. Toxicity was lower in lice exposed orally to IPM (56.0%) than by direct contact with a treated substrate (90.4%). Direct contact gave signs of desiccation, oral exposure did not. A cuticular route of action is indicated. An IND has been submitted to FDA and Phase II clinical trials start in April 2004.

Development of new microsatellite markers in the New World screw-worm *Cochliomyia hominivorax* (Diptera: Calliphoridae)

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Microsatellites are a highly informative class of DNA markers, consisting of tandem repeats of 2-6 bp distributed throughout eukaryotic genomes. Microsatellite loci stand out as codominant and multiallelic markers with high polymorphism and expected heterozygosity value. Due to these characteristics, they were the molecular markers chosen to continue our efforts to investigate the genetic variability and population structure of the New World screw-worm (NWS) *Cochliomyia hominivorax* (Coquerel). This species is one of the most important parasitic insect pests in Neotropics, causing myiasis and, consequently, significant losses to livestock industries in these regions. In order to generate new microsatellite markers for NWS, an enriched genomic library for microsatellite inserts was constructed. Of 56 clones inserts sequenced, 30 had a microsatellite and of these, 10 sequences with repeats of di-, tri-, tetra- and hexanucleotides were selected. Primer pairs were generated for the ten selected loci and eight of them were successfully amplified in NWS DNA samples. Polymorphism of each locus will be evaluated and the informative markers, in combination with ten polymorphic microsatellite markers previously described, should provide high resolution for assessing the fine-scale genetic structure in NWS.

Genetic structure and diversity in new world screw-worm *Cochliomyia hominivorax* (Diptera: Calliphoridae) populations from Uruguay revealed by microsatellite markers

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The New World screw-worm (NWS), *Cochliomyia hominivorax* (Coquerel), is one of the most important parasitic insect pests that cause invasive myiasis in warm-blooded vertebrates. Because of the substantial economic losses to livestock rearing caused by this pest throughout the neotropics, an international effort has been involved in a program to eradicate the NWS from endemic areas of Central and South America and to prevent invasions into screw-worm-free areas. It is crucial to characterize the genetic variability of target populations in order to maximize the effectiveness of the eradication program. Ten polymorphic microsatellite loci previously isolated from an enriched library constructed for *C. hominivorax* are currently being used to investigate the genetic variability and population structure across its current geographic distribution. Preliminary results obtained from Uruguayan populations indicated a considerably high genetic variability. Statistical analysis indicated that most of the genetic variation is contained within populations, while low levels of differentiation were found between them. The investigation into the genetic variability and population structure of NWS from Central and South American populations should provide fundamental information for future eradication strategies.

Isolation of microsatellite loci in the blowfly *Chrysomya chloropyga* (Diptera: Calliphoridae)

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The species *Chrysomya chloropyga* (= *Chrysomya putoria*) (Wiedmann) is a synanthropic blowfly native from African regions. The adults feed and breed in carrion, feces, urban garbage and carcasses. Due to these biological features, *C. chloropyga* have great medical-veterinary importance acting as a vector of pathogens and is considered of potential forensic importance. This fly was probably introduced into Brazil around 1975 with the influx of Angolan refugees. Since then it has spread rapidly in Brazil and other countries of South America affecting the ecological scenario of the native Calliphoridae fauna. Genetic studies based on mitochondrial DNA revealed insufficient polymorphism at a fine scale. Since microsatellites are one of the most powerful markers for studying inter- and intrapopulation relationships, our goal is to develop a set of polymorphic microsatellite markers for *C. chloropyga*. A genomic library enriched for the motifs poli-AC/TG and poli-AG/TC was constructed. We are currently screening this library for microsatellite-containing clones. Of 54 clones inserts already sequenced, 26 had a microsatellite and 12 are useful sequences for designing primers. After polymorphism evaluation, these loci should contribute to population dynamics studies and for understanding the evolutionary context of the introduction of this species in the New World.

Survey of new world screw-worm *Cochliomyia hominivorax* (Diptera: Calliphoridae) for the presence of a point mutation associated with esterase-mediated organophosphate resistance

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The *Cochliomyia hominivorax* (Coquerel) is the main agent of primary myiasis in the South America, generating severe economic losses to livestock industry. In Brazil, its control has been done by the application of insecticides, but the inappropriate use of these chemical agents can select resistant flies. It has been shown in another species of the Calliphoridae family, *Lucilia cuprina*, that a Gly137®Asp substitution in the enzyme carboxylesterase (E3) converts its typical activity to an organophosphate (OP) hydrolase activity resulting in the development of OP resistance in this species. Preliminary efforts to isolate and characterize the E3 gene in *C. hominivorax* using primers previously described for *L. cuprina* resulted in a gene fragment of ~200 bp containing the site of the Gly137®Asp mutation. This region showed high similarity to the orthologous region in *L.*cuprina. The intron position is conserved, however its physical size and nucleotide sequence are not. The partial exon sequence differed from that in *L.cuprina* at 2 nucleotide sites, but no amino acid substitution was predicted. Further analysis will be conducted to determine the association of this point mutation OP resistance in *C.hominivorax* strains. This characterization should allow the identification of resistant individuals, enabling more effective control strategies.

Evaluation of prepared Thai plant-based repellents against mosquito bites under field condition

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The experimental repellent products in the gel dosage form contained 20% clove oil (*Syzygium aromaticum*) and 10% clove plus 10% makaen (*Zanthoxylum limonella*) oil mixture were evaluated theirs efficacy against daytime- and nighttime-biting mosquito comparing with 20% deet (di-methyl benzamide) under field condition using a human bait method. One gram of the product was applied evenly from knee to the ankle of each leg. Shorts and shoes were worn to standardize the exposure area. Other exposed untreated parts of the body were protected against mosquitoes attack by wearing a jacket with hood and gloves, covering the thighs with a plastic sheet. For daytime-biting, 20% clove showed complete protection for 4.0 h and gave 95.7% repellency after 5 h application, whereas 10% clove plus 10% makaen oil mixture and 20% deet provided only 86.8 and 82.7% repellency after treatment, respectively against *Aedes aegypti*. For nighttime-biting, the 3 developing repellents yielded equally excellent (average 97.1% repellency for 5 h against the predominant *Culex quinquefasciatus* and *Mansonia uniformis* but they gave 89.0% repellency against *Cx. tritaeniorhynclus* and *Cx. gelidus*. This finding demonstrates that the repellency of 20% clove oil and 10% clove plus 10% makaen oil mixture in the gel dosage form are possible used against various different mosquito species.

Host grooming efficiency and its influence on the distribution of cat fleas, *Ctenocephalides felis*

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Grooming is usually supposed to be the main factor to affect the final distribution of ectoparasites on hosts, that is, the distribution of ectoparasites can be changed when the grooming ability is refrained or can be different between those on the good and poor groomers. Therefore, in this study we compared the distribution of cat fleas, *Ctenocephalides felis* (Bouché)(Siphonaptera: Pulicidae), on the cat with good or poor ability of grooming and the cat wearing an Elizabethan collar or not. As expected, the results show that cat's grooming efficiency against fleas was significantly varied among individuals and greatly reduced by wearing an Elizabethan collar. However, the effect of grooming efficiency on the distribution of fleas on the host body is not significant. Regardless of cat's grooming efficiency, mean percentages of fleas detected at 24 or 96 hours after initial infestation were always highest on the neck (52 – 71.67%) among body regions (other regions ranging from zero to 30%). Fleas seem to prefer the anterior regions of cats including the head, neck, and front back. Moreover, male (60.17%) and female (61.18%) fleas have the same trend to move to the cat's neck.

Impact of anthelmintics on the growth of dung beetles and dung breeding flies in Japan.

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Most of anthelmintrics like an Ivermectin is excreted in the feces as a mixture of parent compound and metabolites. So, the use of anthelmintrics in livestock has potential impact on nontarget invertebrates, for example, dung beetles, flies, that depend on dung pats and contribute dung degradation in grassland. This study examined the impact of the anthelmintics residue on the dung degrading fauna in grassland. Dung samples were collected on 1, 3, 7, 14, 21, 28 and 35 day after pour on and bioassayed using the dung beetles and flies. Ivermectin residues attained maximal concentration from 1st to 3day after pour on followed by a linear decline to 35day. Dung pats collected 1 to 14days after treatment with ivermectin were lethal to larval development of native dung beetle, *Onthophagus lenzii* and native dung breeding flies, *Musca hervei*. Dung collected 1 to 3days after treatment was lethal to larval development and pupation of native dung beetle, *Aphodius elegance*. In field experiment using emergence trap, number of emerged flies were 3,655 (18families) in control dung and 2,234 (16 families) in treated dung.

Investigation of alternative insecticides for the treatment of bed nets and for indoor residual spraying to control *Anopheles* mosquitoes.

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There is an urgent need to investigate alternative insecticides for the control of pyrethroid resistant *Anopheles* mosquitoes, vectors of malaria. Resistance to DDT and pyrethroids among *Anopheles* (especially *gambiae* and *funestus*) species has been reported in western, southern, and eastern Africa. Alternative insecticides with novel modes of action remain may have potential for use on nets or indoor residual sprays. The Gates Malaria Partnership at LSHTM is working with WHO and industry to identify new actives. Bioassay tests were carried out using new insecticides including a phenylpyrazole (fipronil), an organophosphate (chlorpyrifos methyl, "Reldan"), an oxadiazine (indoxacarb) and a pyrrole (chlorfenapyr). Some of these compounds were tested in mixtures to investigate their possible synergistic interaction with pyrethroid insecticides. Laboratory studies with fipronil on netting have shown some irritant effect against *An. stephensi* and some synergisms between fipronil and deltamethrin. Different formulations of Reldan sprayed on various substrates (mud, plaster and wood) to investigate the efficacy of the formulations for indoor residual spraying (IRS) showed good residual performance up to 6 months especially on plaster. On the treated nets, Reldan performed well against wild populations of pyrethroid resistant mosquitoes. Indoxacarb and chlorfenapyr showed a slow acting effect in terms of mortality and KD but indoxacarb was better on netting when mixed with deltamethrin.

Evidence supports two cytologically different races of ***Anopheles pullus*** **in Korea**

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Despite the medically-importance of anopheline mosquitoes as vectors of Korean vivax malaria, differentiation among Korean anopheline mosquitoes are obscure and difficult by traditional morphological taxonomic criteria. To verify the species status of two ambiguous species of *An. pullus* and *An. yatsushiroensis* (*An. pullus form yatsushiroensis*) mosquitoes from Korean, we established isofemale lines and investigated their genetic relationship by metaphase karyotypes, hybridization experiments, DNA sequence comparisons of rDNA ITS2 and mitochondrial COI and COII. Two isofemale lines showed different chromosomal shapes in sex chromosomes of X and Y, the reciprocal crosses between the two isofemale lines, however, provided viable progeny with completely synaptic salivary gland polytene chromosomes. DNA analyses also strongly supported the conspecificity of these isofemale lines. Based on these evidences, we conclude that two *An. pullus* forms which differ distinctively in morphological, cytological and ecological traits remain in interfertile status.

Incompatible hybridization between two sympatric species of *Anopheles sinensis* and *Anopheles pullus* (Diptera: Culicidae) in Korea.

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Hybridization tests of laboratory-raised, iso-female lines of *Anopheles sinensis* Form B and *An. pullus* Form A and B were conducted by induced copulation. The three iso-females line were established based on morphological characteristics of wild-caught, fully engorged females and three distinct types of metaphase karyotypes. They were *An. sinensis* form B (X, Y_2), *An. pullus* form A (X_1,Y_1) and form B (X_2,Y_2). The results of reciprocal and back crosses among *An. sinensis* and *An. pullus* indicated that *An. sinensis* was genetically incompatible with *An. pullus*, providing low viability of F_1-hybrid larvae with completely asynaptic of salivary gland polytene chromosomes, sex distortion of adults, abnormal development of ovarian follicles of F_1-hybrid females, and atrophy of accessory glands and testes of F1-hybrid males. The results strongly suggested that these two morphological species exhibited entirely reproductive isolation.

Bidirectional signalling and synaptogenesis: semaphorin1a and the transition from growth cone to synapse.

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Target recognition during which a motile growth cone is transformed to a sedentary synapse is poorly characterized at the molecular level. We have demonstrated in the *Drosophila* giant fiber system that this transition is regulated in part by semaphorin1a (Godenschwege et al., 2003). Sema1a is a trans-membrane member of a family of proteins typically thought of as repulsive ligands that guide growth cone path finding. We have shown that Sema1a is required in both the pre- and postsynaptic cells for normal synapse formation and are using Sema1a as a point of entry to discover the signaling systems that regulate the transition from growth cone to synapse. In the pre-synaptic giant fiber Sema1a functions as a repulsive receptor and when Sema1a is over-expressed in the giant fiber synapse formation is disrupted. This gain-of-function defect can be enhanced by over expression of rac1 and suppressed by expression of the dominant negative rac1 suggesting that the repulsive function of sema1a is dependent on the small GTPases. We will elaborate on the role of rac1 as well other signaling systems that interact with semaphorin1a and are required for synaptogenesis . Finally, a model for the assembly of this giant synapse will be presented.

Bionomics of *Anopheles hyrcanus* in resurgent and endemic foci of malaria in Iran

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An.hyrcanus group is reported from different parts of Iran. This species plays a role in malaria transmission in neighboring countries. The adult and larvae were collected from different parts of Iran using WHO's methods. This species is found in shaded part of rice-lands and irrigation canals and shaded stagnant water in Nikshahr and Pars-Abad and Poldasht districts. Its peak of activity occurs in midnight whereas s area three peaks of biting can be observed, mainly sunset and midnight according to locality. Light trap is able to collect the adult. This species tend to rest indoor when temperature is falling dawn. The peak of its activity is mainly in August. From the field study it was concluded that *An.hyrcanus* population found in different resurgent and malaria endemic area of Iran. Their seasonal activity, biting habitat, larval density, and abdominal condition is varied in different localities. It seems that in some area due to high population density and antropophilicity of this species it may play a role in malaria transmission. At the future, further molecular and biochemical studies will be done to confirm different sibling species of this species as in studied area.

Behavioral responses of *Culex* to oviposition site odors

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Culex mosquitoes, as important vectors of arboviruses in North America, are effectively sampled by gravid female traps baited with infusions such as hay infusion. Using several methods, a wide range of volatile compounds have been identified from hay and grass infusions and dairy effluent. The purpose of this study was to evaluate the behavioral responses of gravid *Culex* to these compounds as oviposition site attractants in a dual-port olfactometer assay. Responses obtained to individual compounds and then in binary and/or tertiary blends were compared to hay infusion. The efficacy of lures incorporating these mixtures were also evaluated in the olfactometer and compared to hay infusion standards. These results contribute to the development of standardized lures for gravid *Culex*.

Effect of fire on recovery and identification of entomological evidence at a homicide scene

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Insects are valuable in determining elapsed time since death in a homicide. However, the killer may attempt to disguise the crime by setting fire to the crime scene. Experiments were conducted to determine the effect of arson on entomological evidence. Three freshly killed pigs (*Sus scrofa* L.) were placed in separate rooms in a house in Edmonton, Alberta. Extensive blow fly (Diptera: Calliphoridae) colonization occurred, and the remains were allowed to decompose to the advanced decay stage. At the time of the fires, large numbers of larval Calliphoridae, pupae and empty puparia, as well as teneral flies were present. Four successive simulated arson fires were set then extinguished. The first fire was ignited using gasoline poured on the carcass, the second used common household accelerants such as newspaper, the third was set in the basement and the final fire involved the entire house. After each fire was extinguished, the carcasses were examined for entomological evidence. In all, cases, enough entomological evidence was recovered after each fire, including the final destruction of the house, to allow an entomologist to analyze the evidence and to determine elapsed time since death. In several cases, live insects were recovered.

Novel strategies for arbovrius surveillance in response to WNV

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The outbreak of West Nile virus (WNV) in 1999 in Queens, New York USA was an unprecedented public health event on the North American continent. Albeit, the epidemic has stimulated development of only a few novel vector/virus surveillance tools. The VecTest® is the best-known novel technology resulting from the outbreak of West Nile virus. This lateral flow or dipstick test is used to detect West Nile virus antigen in homogenized mosquitoes. The VecTest has also been used successfully to detect viral antigens in extracts of cloacal swabs taken from moribund WNV-infected birds. Another more recent diagnostic test, the RAMP West Nile Virus Test, detects WNV antigen in mosquitoes and birds. This test is based on latex particles that are tagged with fluorescent antibodies to WNV. Sentinel animals have been used to detect the occurrence of WNV. The virus is highly lethal to some birds, such as American crows and blue jays. Field surveillance of WNV activity involves a strategy of systematically collecting and testing dead corvids for WNV infection. Counts of dead corvids have also been used to estimate the intensity of local transmission of WNV.

Weather station ambient temperature corrections in forensic entomology

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Weather station data is used by forensic entomologists to estimate ambient temperatures at body discovery sites. Correlation data from sites may also be collected to correct for site and station differences. This experiment tested the accuracy over time of retrospective weather data correction using linear regression between stations and sites. Temperatures were logged at six hypothetical body discovery sites during a seven day period for which a hypothetical body lay *in situ*, and a further four correlation periods. The accuracy of weather data for hypothetical body *in situ* periods improved after correction in 22 of 24 correlations, however, mean predicted body *in situ* temperatures for sites differed significantly between correlation periods. Predicted temperatures generally rose with time after body removal, which correlated with rising site temperatures accompanying seasonal change. Therefore, practitioners should be cautious in making correlations if weather patterns during correlation differ greatly from those while the body was *in situ*.

Biometrics and behavior in mosquito repellent assays

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Biotic and environmental factors influence the outcome and interpretation of repellent bioassays. Mosquito-related test factors include larval nutrition, carbohydrate availability, age/parity of females, blood engorgement status, and diel blood-feeding patterns. Environmental/human subject-relevant factors are light, temperature, humidity, repellent dose and exposure time, absorption, penetration, evaporation, abrasion, and/or washing of repellent from skin (by perspiration), and the innate repellency of human/animal test subjects. In laboratory tests, cage configuration is critical. With small cage/modular-types of test apparatus (<1000 cm^3), treatment effects may be confounded with "edge effects", because of the position of a feeding port or module, whereas tests repeated using the same human subject comprise pseudo-replication. In large cage (>1000 cm^3) tests, complete protection times are based on the responses of mosquitoes in the upper extreme of the frequency distribution for repellent tolerance, rather than on the average population response; large cage techniques are not useful for determining the Estimated Median Dose (ED_{50}), or other percentiles of interest. Variation in repellent assays can be minimized by rearing and testing mosquitoes in a standardized environment, by choosing appropriate sample sizes, by selecting a biologically-relevant assay method, and by careful and diligent oversight of test proceedings.

Vector and viral genetic determinants of epidemic dengue and dengue hemorrhagic fever in Mexico

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To elucidate vector determinants of dengue transmission in Mexico, ~1,600 *Ae. aegypti* collected from throughout North America and individually phenotyped for dengue 2 (DEN2) virus midgut and disseminated infections. DNA from each of these mosquitoes was archived for association mapping of candidate genes that condition vector competence. Dramatic differences in vector competence in the *Aedes aegypti* populations were demonstrated, and vector competence was correlated with dengue seroprevalence. Studies to reveal the genetic basis of *Aedes aegypti* vector competence identified two candidate genes (Early Trypsin and Abundant Trypsin) that condition dengue virus infection of vectors. RNAi silencing of the Early Trypsin gene resulted in reduced vector competence of *Aedes aegypti*. To determine the epidemiological relevance of these genes, a population genomics approach using association mapping was used to reveal the statistical relationship between genetic polymorphisms and vector competence phenotype in these genes. An association was found between the *Abundant Trypsin* and DEN2 disseminated infection rates DEN2 infection phenotypes in these American *Aedes aegypti* populations. Such information is important in risk assessment for epidemic dengue and DHF/DSS. During these studies, a new DEN2 virus of the American-Asian genotype was introduced in Yucatan State, causing an epidemic of DHF/DSS. This virus is being compared with other DEN2 viruses, which have previously circulated in the Yucatan State, for its ability to infect, replicate in, and be transmitted by *Aedes aegypti* and for its epidemic potential.

Chemical analysis and laboratory bioassays of host odors to identify compounds that attract and inhibit the attraction of mosquitoes

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Using various sampling techniques and gas chromatography/mass spectrometry (GC/MS), a comprehensive chemical profile of host odors can be obtained. The differences in chemicals emanated by different hosts may provide a clue to the structures of compounds used by anthropophilic mosquitoes, compared to those used by zoophilic mosquitoes, for host location. Screening of candidate compounds is accomplished in our laboratory by using a triple-cage dual-port olfactometer. Bioassays of single compounds and blends of compounds have resulted in the development of attractive synergistic binary blends for anthropophilic mosquitoes. Further improvement to the blends has resulted in a three-component blend that is more attractive than some human individuals in laboratory bioassays. During the development of these attractive odor blends, some compounds were observed to inhibit the host-seeking capability of mosquitoes, possibly my masking the attractive odors. Inclusion of these compounds (attraction-inhibitors) in attractive blends, results in a substantial decrease in the ability of mosquitoes to locate the attractive odor source in the olfactometer. Laboratory bioassays are being used to develop a system that demonstrates the possibility of concealing a host with attraction-inhibitors and driving mosquitoes to choose an attractant blend over a live human or animal host.

Sexual dimorphism of antennal gene expression in the malaria vector *Anopheles gambiae*

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To obtain a better understanding of the olfactory processes that allow mosquitoes to identify human hosts, a molecular study has been performed to identify and characterize molecules in the olfactory signaling pathway of the African malaria vector *Anopheles gambiae*. cDNA libraries from antennae of females and males were established, from which a collection of cDNAs encoding odorant binding proteins (OBPs) and other novel antennal proteins were isolated and characterized, representing various families of putative carrier proteins with homologues in other insects. Using filter array hybridizations and quantitative RT PCR, regulation and gender specificity of expression of these genes was investigated. Although male and females appear to express the same olfactory genes, significant differences in steady state levels of some of these putative carrier protein genes were detected between the sexes and after blood feeding in females. Results of microarray experiments of gender-specific expression of olfactory carrier proteins and receptors will be reported. Our ultimate goal is to identify and characterize molecules in the odor reception pathway of this mosquito vector species with the intent of facilitating the development of better methods to interfere with the mosquito-host interaction.

Study of decomposition rates of pig carcasses and viability of *Calliphora vicina* in the UK climate

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The estimation of post mortem interval (PMI) in a country such as the UK which experiences fluctuating temperatures, both seasonally and daily, is considerably more complicated than in those countries with fairly constant temperatures. In order to assess the effects of the different seasons on decomposition rates and insect species composition, piglet carcasses were put out at The Natural History Museum in Central London. Significant differences were recorded from carcasses put out at different times of the year, and between those laid outside on a roof, and those inside the building, where insects had limited access to the carcasses. Although oviposition by the primary invaders occurred at the same time on both the indoor and outdoor carcasses, the rate of development of *Calliphora vicina* on the outdoor carcasses was significantly slowed down due to the cold temperatures experienced, thus limiting the decomposition rate considerably. Further laboratory based studies were carried out to assess the effect of cold temperatures on the different life stages of *Calliphora vicina*.

Molecular basis of resistance to pediculicides

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Head and body lice develop resistance to pediculicides through mutations of insecticide target sites, such as the voltage-gated sodium channel, and increased activity of detoxification enzymes such as oxidases and esterases. The difficulties of conducting bioassays on field-collected lice make molecular analysis of insecticide resistance an especially attractive option. Here are described the molecular details of resistance that has appeared in lice together with a discussion of ongoing work to develop methods to detect and assess these mechanisms through molecular analysis. A useful approach to measuring all of these mechanisms is fluorometric PCR. This method allows detection of resistance mechanisms based on point mutations at the insecticide target site, resistance gene amplification, or increased expression of resistance genes. Knowledge of resistance gene sequence is a prerequisite in the development of these methods. Our knowledge to date of louse resistance gene sequences is summarized. The same molecular methods that serve to detect resistance are also central to the determination of population genetic relationships of the resistance alleles. We discuss our present state of knowledge of louse molecular markers and the louse genome as they relate to insecticide resistance.

Environmental change and lyme disease risk

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An understanding of the influence of environmental change on Lyme disease in North America is a fundamental component in assessing changes the spatial distribution of human risk. Because disease risk may be influenced both by large-scale climate change and local-scale landscape modification, we developed a multi-scale approach to dissecting the influence of environment. First, we used a climate suitability model for the tick vector *Ixodes scapularis* to examine the potential effects of global climate change on future Lyme disease risk in North America. The model reveals a significant expansion of *I. scapularis* north into Canada and central United States as well as a retraction of the vector from southern United States. With higher resolution remotely sensed data, we evaluated the impact of local-scale landscape structure on both entomologic risk and human exposure. Landscape fragmentation resulting from suburban development was compared to both human and tick surveillance data from Connecticut and showed that landscape mosaic plays a significant role in defining disease risk. In spite of higher tick densities, human incidence of Lyme disease was found to be lower in fragmented contexts. The synthesis of such environmental models will allow for improved predictions of Lyme disease emergence.

Influence of the structure of host-odor plumes on the orientation maneuvers of flying mosquitoes

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Host finding in mosquitoes is typically subdivided into two sequential stages: attraction at long range that is induced by host-released odors, principally assumed to be carbon dioxide. Orientation at close range is assumed to be regulated by the interactions of carbon dioxide with other host odors, humidity, heat, and visual cues. In our work with *Aedes aegypti* in a wind tunnel, upwind fight along the carbon dioxide plume required the sequential encounter with filaments of carbon dioxide, as would normally occur in a plume diffusing in wind. A single brief encounter with a filament of carbon dioxide odor induced a surge toward upwind. A relatively homogeneous cloud of carbon dioxide, in contrast, activated flight, but did not stimulate upwind movement. Conversely, orientation to human skin odor seems to require a longer term encounter than occurs with exposure to an odor filament. The threshold for response to skin odor is itself modulated by an encounter with carbon dioxide: a brief exposure to carbon dioxide lowers the threshold of response to skin odor by five-fold. Together the interactions of carbon dioxide and skin odor explain several features of host orientation, and provide support for the hypothesis of sequential stages of odor utilization.

Development of lure for the dengue mosquito

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Instead of proceeding with the traditional isolation and identification from hosts of individual mosquito attractants, ISCA Technologies, Inc. created compositions which mimic the skin of the specific vertebrate hosts. Most mosquitoes apparently exhibit some level of host 'preference', i.e., they do not feed indiscriminately on any vertebrate host. In some studies host-feeding patterns within a closely related species complex is odor mediated and was attributed to differential response to CO_2 and other skin odors. The actual bait, created through a process patented by ISCA, provides the parasitic arthropod (host seeking *Aedes aegypti*) a much more complete "olfactory sensation" associated to a specific "host" (human) than any other attractants on the market. Traps lured with this bait containing artificial "sensory correct" profiles of human hosts trigger rapid responses from females *Ae aegypti* inducing the full set of host seeking and feeding behaviors, from activation to imbibing. The current ISCA prototype trap has proven to be superior to the Fay-Prince and Wilton traps and other commercial traps set-up in competitive laboratory trap tests. Laboratory studies indicate the capture rate of this prototype baited with this new lure closely approximates the rates obtained from human biting counts for the same mosquito density.

2-Oxocarboxylic acids elicit landing responses from female *Anopheles gambiae*

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Landing and probing responses were elicited from female *Anopheles gambiae* in wind tunnel bioassays by 2-oxocarboxylic acids. The most significant responses were elicited by 2-oxopentanoic acid although 2-oxo-butanoic, 2-oxo-3-methylbutanoic, 2-oxo-3-methylpentanoic, 2-oxo-4-methylpentanoic and 2-oxo-hexanoic acids also elicited responses. 2-Oxo-propanoic (pyruvic), 2-oxo-octanoic, 2-hydroxy-propanoic (lactic) and 2-hydroxy-pentanoic acids were inactive. The response to 2-oxopentanoic acid was dose and temperature dependent with 30 µg and body temperature (34^{o}C) eliciting the most significant responses. Responses decreased over time but this was thought to be due to evaporation. Chemical analysis of a suitably derivatized human sweat sample confirmed the presence of pyruvic, 2-oxo-3-methylbutanoic, 2-oxo-3-methylpentanoic and (*E*)-oxo-4-methylpentanoic acids. The three methyl-branched acids, previously identified in human body fluids, are metabolically linked to the amino acids valine, leucine and isoleucine *via* a reversible trans-amination. However, the most active compounds, 2-oxopentanoic and 2-oxohexanoic acid were not identified in human sweat. Taken together the research suggests that C_4-C_6 2-oxocarboxylic acids represent a class of compounds that provide female *An. gambiae* with close range, host-specific odour cues.

Distribution of blood feeding within the subgenus *Inseliellum* (Diptera: Simuliidae)

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Simulium (*Inseliellum*) *buissoni* in Nuku Hiva, Marquesas Islands, Polynesia, has been referred to as the "scourge of the Pacific". It is a sufficiently serious blood feeder on humans that it resulted in the administrative centre of the Marquesas being moved from Nuku Hiva, south to Hiva Oa. *Simulium buissoni* is also a serious bird biter and attempts to eradicate this black fly were made in the early 1990's. *Simulium* (*I.*) *gallinum,* distributed on other Marquesas Islands, however, feeds only on birds. Distribution of blood feeding within *Inseliellum* will be discussed in light of a reconstructed phylogeny for the subgenus. Also discussed will be the "Miracle of the Nono" from the 1930's, where it is reputed that serious biting of humans by black flies on the southern Marquesas Islands, ceased ostensibly, overnight.

Evolution of blood-feeding behavior in black flies (Diptera: Simuliidae)

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Phylogenetic-, paleontological-, and host specificity data are combined to reveal patterns of blood feeding behavior among black flies. The fossil record reveals that simuliids originated before the radiation of birds and mammals, suggesting that other warm-blooded hosts were exploited by the earliest members of the family. Ornithophily probably evolved independently in the Prosimuliini and Simuliini, with numerous, independent, reversals to mammalophily in the latter tribe. Although host-switching events in black flies have been predominantly from ornithophily to mammalophily, there is a least one example of the reverse among certain members of the *Simulium* (*s.str.*) *slossonae* species-group. Obligatory autogeny (i.e., females that lack biting mouth parts), is a derived trait that evolved numerous times independently from both mammalophilic and ornithophilic progenitors. Such non-bloodsucking species, which constitute only about 2.5% of the world species, live predominantly in high latitude or high altitude environments. Tarsal claw morphology has utility for inferring host preference among anautogenous black flies; however, the current practice of coding claws as either "simple" or "bifid" does not exploit the full panoply of variation present in this character system. Detailed comparative studies are needed in order to identify useful characters for species recognition and phylogenetic studies.

Landscape epidemiology of malaria in Tumkur (Karnataka), India, as monitored by satellite remote sensing

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Study was conducted in selected villages of 3 Primary Health Centres of Tumkur (Karnataka)with varying endemicity to find out the landscape elements critical to malaria endemicity at village level using satellite remote sensing.. Ground data on the presence of breeding habitats, monitoring of larval density, adult *Anopheles culicifacies* (malaria vector) from indoor resting sites and ecological features within 1.5 km radius from the centre of each village were generated in 2001 and 2002. Data of Indian Remote Sensing satellite with 23.8 metre and 5.8 metre resolution respectively were procured for the dates of ground survey and analyzed for generation of images and statistics of land use features within a buffer zone of 1.5 km around each village. Delineation of tanks, streams, ponds, marshy areas, the main breeding habitats of *An. culicifacies* was possible by Remote sensing by merging LISS III and PAN products. The breeding habitats in the vicinity of human settlements within 500 metres were found more productive for *An. culicifacies.*

Quantification of land use features revealed that extent of water bodies, permanent vegetation cover, less barren/scrub and rocky areas were associated with high malaria endemicity. Identification of villages into high/low risk of malaria is possible leading to village level stratification for development of village specific selective and cost effective spraying strategy for integrated vector management programme for targeting malaria control.

Aedes aegypti behavior in manipulated domestic environments in Thailand

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Aedes aegypti, a low-density mosquito, lives in or near human dwellings, feeds frequently and specifically on people, and efficiently transmits dengue viruses. Field experiments with wild mosquitoes are risky in endemic areas and population augmentation to increase sample sizes causes ethical concerns. We circumvented both issues by screening the entire house and yard of rented homes in order to safely conduct relatively natural behavioral experiments using females collected in the village as pupae. Host size, location, and orientation effects on feeding were studied using 4 rotated subjects. Released mosquitoes (~25%) were recovered using pack-pack aspirators and the origin of their blood meals scored by DNA fingerprinting. There was strong correlation between host size and feeding with the largest individuals consistently feeding the most mosquitoes. *Ae. aegypti* fed most often on people inside house but body orientation appeared unimportant. Timing of oviposition and the impact of container size on egg number were evaluated in various sized jars lined with germination paper. Papers were changed at regular intervals and eggs counted. Most eggs were laid in late afternoon. Larger diameter containers consistently received more eggs whether located inside or outside. Indoor females preferred containers without preexisting eggs.

Behavioural responses of gravid *Aedes aegypti* (Diptera: Culicidae) to synthetic oviposition attractants identified from grass infusions volatiles.

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Grass infusions have been used as oviposition attractants and/or stimulants for enhancing ovitrap efficacy. However, the infusion procedures are very time-consuming and extremely labour-intensive. Thus, the volatiles released from grass infusions that attract gravid mosquitoes *Aedes aegypti* to ovitrap were identified by coupled gas chromatography-electroantennogram detection (GC-EAD), coupled gas chromatography-masspectometry (GC-MS), electroantennography (EAG) and field trapping. Six candidate compounds were identified and evaluated in the field and in laboratory. Electrophysiological studies showed that antennas of gravid mosquitoes are sensitive to all synthetic compounds but only few individual compounds elicited behavioral responses within a dual port olfactometer. Laboratory and field studies showed that the lures and dispensers are still attractive after 30 days old in the environment. Video recording analysis of the behavior response of gravid *A. aegypti* to adult mosquito trap (MosquiTRAP^{â}) baited with the oviposition lure (Atr*Aedes*^{â}) showed that chemical and physical structures of odor plumes have great influence on the trap catch performance. At the moment, the MosquiTRAP^{â} and the Atr*Aedes*^{â} are being evaluated for *A. aegypti* national monitoring program in Brazil.

Laboratory and field evaluation of mosquito repellents in Australia

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This paper will review laboratory and field studies conducted in Australia and Papua New Guinea to evaluate mosquito repellent formulations. The response of *Anopheles* spp. in the field to *N,N*-diethyl-*3*-methylbenzamide (deet) is variable, and several species are tolerant of current formulations. In contrast, deet repellents are quite effective in providing protection against Culicine mosquitoes in this region. Additional studies on the laboratory and field response of mosquitoes to a comparatively new active ingredient, (1-methyl-propyl 2-(2-hydroxyethyl)-1-piperidinecarboxylate), known as picaridin, will also be presented.

Role of defensin in tick-pathogen interaction

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Antimicrobial peptides are major components of host innate immunity, a well-conserved, evolutionarily ancient defensive mechanism. Infectious disease-bearing vector ticks are thought to possess specific defense molecule that have been acquired in the process of their evolution against the transmitted pathogens. We found a parasiticidal peptide evolved from a common ancestral peptide resembling spider and scorpion toxins from *Haemaphysalis longicornis*, the obligatory vector for *Babesia* parasite transmission. This protein, named longicin, displayed bactericidal and fungicidal properties resembling those of defensin homlogues from invertebrates and vertebrates. Longicin showed a remarkable ability to inhibit the proliferation of merozoites, an erythrocyte-blood stage of equine *B. equi*, by killing the parasites. Longicin was localized at the surface of the parasites, as demonstrated by confocal microscopic analysis. In an *in vivo* experiment, longicin induced significant inhibition of parasitemia in animals infected with the zoonotic and murine *B. microti*. Our findings suggest that longicin is a promising candidate for designing antibabesia therapeutics.

Attractive host odours for mosquitoes: the blend ratio makes the difference

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Mosquito species differ with respect to their host preferences, and some discern not only different species of vertebrate hosts but also different individuals of a given species. Olfactory cues derived from the hosts determine to a large extend the host choice, and it is an intriguing question how these insects accomplish the necessary fine odour discrimination. Emanations from vertebrate hosts consist of complex blends. Attention has focused on the identification of kairomones that guide mosquitoes to their hosts. Here we show that odour blends rather than their single constituents attract the anthropophilic yellow fever mosquito *Aedes aegypti* (L.). Highly attractive blends consist of L-(+)-lactic acid, ammonia, and distinct fatty acids which were essential to maximise the attractiveness of the blend. The mosquitoes' responses, however, depended markedly on the ratio of these compounds. In a direct competition bioassay a distinct blend ratio turned out to be as attractive as an average human hand. When added to natural human skin emanations some compounds also altered significantly the attractiveness of the host. The data show that the blend ratio of a few distinct compounds is crucial for the attraction of mosquitoes to human hosts.

Specific monitoring tools for anthropohilic mosquitoes

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For a better estimation of the risks of mosquito-transmitted disease epidemics, it is vital to quickly assess the density of the host seeking mosquito population. A simple, specific, and effective trapping tool would overcome some of the disadvantages of the currently used methods to estimate the density of mosquito populations. We introduce an new monitoring suction trap with an attractive lure for anthropohilic mosquitoes. The lure releases a cocktail of volatiles that also emanate from human skin. The attractiveness of the mixture strongly depends on the mixing ratio of the compounds. Release rates and blend ratio can be optimised to suit a number of anthropophilic mosquitoes. Although CO_2 is not essentially necessary for the effectiveness of the new attractant, it enhances the catching rate of the blend.
The design of the trap combines the generation of a specific plume shape for an optimum effectiveness of the attractants with visual cues to enhance the catch of the attracted mosquitoes. The trap is optimised for the new attractive blend, easy to produce and simple to use. It can be applied indoors as well as outdoors and is currently tested in the laboratory and in the field in Brazil, Asia, Australia and Europe with a range of mosquito species.

Spatio-temporal risks for lyme borreliosis: examples from Switzerland

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Lyme borreliosis causes health problem for populations in endemic areas in the Northern hemisphere. In the present study, the density of questing *Ixodes ricinus* ticks and their infection with *Borrelia burgdorferi* sensu lato (sl) was examined in areas located North and South of the Alps in Switzerland. From 1999 to 2001 free - living *I. ricinus* ticks were collected on a monthly basis by flagging vegetation in these areas. Density of questing ticks varied greatly between studied areas. *Borreliae* were observed in ticks collected in all investigated sites. However the prevalence of infection differed significantly among areas. Infection prevalence varied from 9% to 40% in nymphs and from 17% to 51% in adults. Adult ticks were significantly more infected (35%) than nymphs (20%). There was no correlation between nymphal density and infection prevalence as well as between adult density and infection prevalence, but there was a correlation between density of ticks and density of infected ticks. During the spring peak of questing tick density, a range of 2-69 infected ticks per 100 m^2 was observed. Five *Borrelia* species were identifed: *B. garinii*, *B. burgdorferi* sensu stricto, *B. afzelii*, *B. valaisiana* and *B. lusitaniae*.

Forensic entomology and artificial intelligence: which is the relation?

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Blowflies of genus *Chrysomya* are so important in forensic entomology to indicate the Post Mortem Interval (PMI). Moreover, the larvae of these insects have an aggregation behaviour when are feeding of the tissues in human and animal corpses which each one influence the other one during this process of feeding and post- feeding, and still after, during the post- feeding larval dispersal searching for pupation sites. The results of this influence is a self-organization in this process which is studied in computer programs (Cellular Automata) using the data from this behaviour and the program produced from these data to be applied in the development of programs of Artificial Intelligence and improve the performance of navigation systems in robots by the environment. These studies were done with about 10,000 larvae of *Chysomya megacephala* and *Chrysomya albiceps* in arenas simulating the environment conditions. Therefore, the study of some biological systems just as the studies in Forensic Entomology could be so important to develop programs in Artificial Intelligence.

Natural repellents against malaria vectors – an evolving operational research agenda in southern Africa

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Personal protection against biting mosquitoes is an important malaria prevention strategy. Insecticide-impregnated bednets and indoor residual house spraying, the main strategies for malaria vector control, provide protection only against indoor feeding and indoor resting malaria vectors. Furthermore, the development and spread of antimalarial drug resistance and insecticide resistance in vector mosquitoes has reawakened interest in repellent use for malaria control. However, currently available synthetic repellents are not affordable to the majority of people living in malaria-endemic countries. Results of qualitative enquiry in Mpumalanga province, a rural malaria-endemic area of South Africa bordering Mozambique lead to identification of three plants, *Lippia javanica* (fever tea), *Pelargonium reniforme* (rose geranium) and *Cymbopogon excavatus* (lemon grass), traditionally applied for preventing mosquito bites. We report on the laboratory and subsequent field evaluation of *Anopheles arabiensis* repellency using water and alcohol leaf extracts from the three plant species. Efficacy results for *L. javanica*, a pleasant-smelling shrub that is locally abundant during the malaria season suggest its utility as a cost-effective personal repellent.

Role of tick lectins in pathogen transmission

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Tissue specific lectin/haemagglutinin activities of both soft and hard ticks have been investigated. Some tick lectins are proteins with binding affinity for sialic acid, various derivatives of hexosamines, and different glycoconjugates. Most tick lectin/haemagglutinin activities are blood meal enhanced, and could serve as molecular factors of self/non-self recognition in defence reactions against bacteria or fungi, as well as in pathogen/parasite transmission. Dorin M, the plasma lectin of *Ornithodoros moubata*, is the first tick lectin purified so far from tick haemolymph, and the first that has been fully characterized. Partial characterization of other tick lectins/haemagglutinins has been performed mainly with respect to their carbohydrate binding specificities and immunochemical features. Few papers have been published on tick lectins so far, and therefore more data is needed to complete the mosaic of knowledge of their structural and functional properties in terms of their role in pathogen transmission. (Supported by the Grant Agency of the Czech Republic No. 206/03/1323.)

A three dimensional pharmacophore model for discovery of new insect repellent compounds

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In pursuit for a better understanding of insect repellent property of molecules we have generated a three-dimensional pharmacophore model using 3D-Catalyst from a training set of eleven diverse insect repellent compounds including DEET. Protection time data for repellent activity of the training set were taken from an earlier published study. Three hydrophobic sites and a hydrogen-bond acceptor site in the molecule appear to be critical for potent insect repellent activity. Calculated stereoelectronic properties such as molecular electrostatic potentials on DEET and some of these molecules in the training set are consistent with our observation. The pharmacophore shows a fairly good correlation (correlation=0.9) between protection time afforded by the compounds in the training set and their predicted protection time. The validity of the pharmacophore model goes beyond the list in the training set and is found to map well on a variety of other insect repellents. By mapping this model on a potent insect repellent compound we have generated a three-dimensional shape based template. A search of in-house database using the pharmacophore template resulted in the discovery of new potential candidate repellent compounds.

Making the case: frequently-contested aspects of forensic entomology evidence

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As forensic entomology evidence is introduced at trial, there are a few predictable aspects that frequently are at the heart of controversy regarding either admissibility or weight. Essentially all of these relate to the argument that, whereas forensic entomology is "good science" generally and applied in the present instance by a qualified expert, that as applied to the facts of the case at bar, the opinion under consideration is not based on reliable data (standards of the U.S. Federal Rules of Evidence or various state codifications thereof). Of those objections related to such "reliable data," the most common related to forensic entomology center around the use of remote temperatures. Few homicide victims are located proximate to weather stations, and fewer still decompose while investigators make measurements. Consequently, in virtually all instances, it is necessary to analyze the thermal development of insect species in the context of temperature data obtained during routine operations of weather stations that typically are located at some distance remote from the site where the decedent was found. Whether these data are analyzed as accumulated degree-days (ADD) or degree-hours (ADH), the same objections are continually raised in court: that the data do not accurately reflect conditions at the decomposition scene (because of uncertainty due to distance, differences in altitude, eccentricities of air flow, cooling effects of adjacent water bodies, heating effect of bright sunlight, or similar concerns). Generally, such objections can be forestalled by simple temperature measurements verifying concordance between the weather stations and body site. Related weaknesses include arithmetic errors in tabulating thermal units, misapplication of ADD to calculate ADH, failure to understand how ADD/ADH are applied in the context of existing dipteran developmental data, failure to consider base temperatures below which development essentially ceases, failure to consider the possible effects of "maggot mass," or putative failure to consider the effect of unique drying conditions on corpses. The forensic entomologist should analyze cases in the context of such potential arguments and be prepared to defend conclusions. Curiously, allegations regarding possible misidentification of insect species are seldom raised.

Blood feeding behavior of west nile virus mosquito vectors in Northeastern USA

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Control and prevention strategies for West Nile virus (WNV) can only be optimized once the most important human vectors have been identified. Although enzootic vectors of WNV have been identified, the epizootic human and horse-feeding vectors are not clear. A three-year study was conducted to investigate the host-feeding patterns of mosquitoes in areas where humans are active and avian reservoirs are nearby. Resting collections, using specially designed vegetation aspirators, were conducted in Upstate New York and New York City. After identification to species, engorged abdomens were analyzed using antibody sandwich ELISA to identify host blood sources. Blood meals also were tested with the precipitin method against antisera for 20 different species of mammals, birds, and reptiles. More than one host in a blood meal was detected in a small number of samples. These results provide a picture of the temporal and geographic variation in feeding patterns of mosquito species commonly included in surveillance programs as well as those that have not been considered important vectors of pathogens in the Northeast USA. The greatest frequency of human blood feeding was found in *Ochlerotatus triseriatus, Oc. aurifer, Aedes cinerius, Oc. fitchii/stimulans, Culex restuans, Cx. salinarius, Cx. territans, Ae. albopictus* and *Oc. japonicus.*

Going global: Taking molecular forensic entomology the next step

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In recent years, the importance of DNA-based techniques for application to the identification of insects, particularly blowfly larvae (Calliphoridae: Diptera) has been widely established. Research has frequently been locality specific, considering the distinction of different species within isolated populations or groups of populations within a small area. While small scale studies are beneficial in development of protocols for accurate molecular identification of insects in specific localities, a global approach to molecular study of forensically significant species facilitates consideration of such issues as species status. This study considers genetic variation in the cytochrome oxidase I (COI) encoding region of mitochondrial DNA (mtDNA) from calliphorids collected throughout Southern Africa and Australia. Alternate regions of DNA are explored for their value in providing an insight to intraspecific variation within several cosmopolitan species. Flies from New Zealand, the United States, Thailand, Taiwan, Brazil and Italy are considered, in addition to Australian and southern African individuals. The study indicates the potential of DNA to provide not only a means of more efficient and accurate identification of insects, but also to analyse the genetic processes occurring within some species, and potential applications and pitfalls for forensic entomology.

The potential for traditional plant-based insect repellents & fumigants as a means of reducing malaria in remote areas

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Insect repellents and mosquito coils are widely accepted as beneficial for travelers in regions of vector-borne disease transmission. However, their efficacy at preventing malaria has never been established and is one reason why such methods have never been extended into disease control programmes. Isolated communities in remote regions are often at increased risk of malaria due to problems in provision of even basic malaria control programmes. For example, *P. falciparum* cases in China are increasing dramatically among minority peoples living in the border regions of Laos, Myanmar and Vietnam, or among migrant forest workers in the Bolivian Amazon. In such areas, particularly where there is an abundant floral habitat such as the SE Asia rainforests or the Amazon, there often remains dwindling historical knowledge of traditional plant-based anti-mosquito measures. By working with such communities it may be possible to identify and develop a means of disease control at the individual, household or village level. Such methods, if effective, could be both cheap and sustainable if local plant species are identified as having repellent, fumigant or insecticidal properties which could be exploited. Surveys of plant use among 25 minority peoples in China are reported and the potential for malaria control assessed.

Incidence, prevalence, and seasonality of *Choanotaenia* and *Raillietina* poultry tapeworms in arthropod and vertebrate hosts

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Confined animal production facilitates arthropod transmission of disease agents by increasing host-vector interactions, fostering sustained exposures, and providing optimal habitat. Poultry tapeworms, *Choanotaenia infundibulum* and *Raillietina cesticillus*, infest domestic and wild fowl throughout the world. Research at the University of California demonstrated that two insect species collected from caged laying hen operations, the hide beetle, *Dermestes maculatus*, and the house fly, *Musca domestica*, were naturally infected with tapeworm cysticercoids. Tapeworm incidence and prevalence fluctuated independently of host populations, with no distinct seasonal pattern. Extrinsic incubation was shown to be temperature-dependent, with development to infective cysticercoids requiring only 5 days at 33C. As husbandry practices change to optimize facilities for food animal productivity, conditions conducive for arthropod populations simultaneously develop. As intensive production increases, infestation rates can be expected to climb as well.

The sonic web trap for management of stomoxys calcitrans around horses

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The Sonic Web (SW) is a trap that is attractive to nuisance flies, particularly the stable fly, *Stomoxys calcitrans*. The trap has a sound component, the heartbeat of a dog, as well as a visual component that draw flies to the trap where they are captured on a replaceable sticky surface. Traps were first evaluated in pairs to determine the role played by the sound. Traps were placed approximately 100 m apart on a horse farm and trap 2 always captured more stable flies than trap 1. The fewest flies were captured when sound was either off or on in both traps. The most flies were captured when sound was on in trap 1 and off in trap 2. Results indicated that several traps could be used in tandem with the sound turned on in only one key trap with spacing at 100-m intervals. SW traps captured significantly more stable flies that the standard Olson trap. When SW traps were placed along the 4 sides of a paddock containing a horse, traps intercepted 90% of the flies entering the paddock before they fed on the horse. SW traps could be an important part of an integrated stable fly management program.

Issues in the control of sheep lice (*Bovicola ovis*)

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Sheep lice are among the three main parasitic diseases of sheep in Australia. Historically sheep lice have been controlled by government regulation and the application of chemical insecticides. However, where regulatory powers still exist they are now seldom exercised and there is increasing sensitivity to the use of chemicals in major wool markets, amongst woolgrowers and in society at large. Woolgrowers face continuing cost of production pressures and the effectiveness of some widely used products has been compromised by the development of resistance. These constraints necessitate a more integrated approach to control. Barriers to change include poor knowledge of the biology of *B. ovis*, difficulty in detecting lice in the early stages of an infestation, lack of availability of alternative control methods and sociological factors. These issues are explored and parallels awn with human head lice control.

Regulation of head lice treatments in Australia

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Head lice treatments in Australia are regulated by the Therapeutic Goods Administration (TGA) via a two-tiered system. Head lice treatments containing herbal ingredients identified as "low risk" (e.g. melaleuca oil) require **listing** on the Australian Register of Therapeutic Goods (ARTG) These products do not undergo efficacy evaluation prior to listing, although sponsors are required to hold evidence to support label claims. Treatments making "high" level claims, or containing ingredients such as malathion, permethrin or pyrethrins require **registration** on the ARTG, involving evaluation of safety, efficacy and quality of the goods. However, a number of currently registered products were available before the onset of the listing/registration process in 1991. Products available before this time may not have been evaluated, although the potential for the TGA to request evaluation of these "grandfathered" products exists. The regulation of head lice treatments in Australia was reviewed in 2002/2003. A number of recommendations resulted from the review including specific recommendations about label claims and the level of evidence required to support registration or listing of a new head lice treatment product. The recommendations take into account the practical requirements of sponsors and the limitations of the current two-tiered regulation system.

A new device for recording host-seeking rhythms of mosquitoes using a photoelectric sensor

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A new automatic device capable of recording the host-seeking behavior of mosquitoes was designed using a photoelectric sensor. The host-seeking rhythms of several mosquito species were recorded in the laboratory. The use of CO_2, in addition to heating and contrast of black and white color, was essential to the functioning of the present recording device. The diel rhythms of non-blood fed *Aedes albopictus, Ae. aegypti, Culex quinquefasciatus, Cx. tritaeniorhynchus*, and *Anopheles stephensi* females recorded by the device corresponded closely with the flight activity levels, as measured by human landing counts and light traps, observed in the field for the same species. This simple automatic recording device also proved to be useful for evaluating the natural host seeking behavior of field-collected mosquitoes.

Astounding success in controlling *Aedes aegypti* in Vietnam using a new strategy

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From 1989, Vu Sinh Nam and Brian Kay have worked together with a team of collaborators including John Aaskov and Truong Uyen Ninh to deliver community based *Aedes aegypti* control using predacious copepods, the programs being managed by the Australian Foundation of Peoples of Asia and the Pacific. The four elements of the new strategy are (1) vertical and horizontal approaches to build up community understanding, (2) quantitative sampling methodologies designed to prioritize control, (3) inoculation of locally acquired *Mesocyclops* into large water storage containers, and (4) vector control activities at community level. By 2003, *Aedes aegypti* has been eliminated from 42 of 46 communes in north and central Vietnam, involving the protection of some 400,000 people. This is a world first achievement as no insecticide was used. The implications of this model are exciting as this approach is applicable to many countries where water has to be stored for survival.

Seasonal studies on the influence of clothing and wrapping on carcass decomposition and arthropod succession

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Six pig (*Sus scrofa* L.) carcasses, three wrapped and clothed carcass and three wrapped and naked, were sampled i) daily ii) after five days and iii) after ten days. Two more carcasses, one naked and not wrapped, and one clothed but not wrapped were sampled daily as controls. Differences in decomposition between the clothed or wrapped carcasses included the distribution and size of maggot masses and significant delay in drying out of the carcasses for all seasons. The maggot masses were primarily dominated by *Chrysomya marginalis* (Wiedemann) and *C. albiceps* (Weidemann), in autumn and summer. *Chrysomya chloropyga* (Weidemann) and *Calliphora vicina* Robineau-Devoidy in winter, with *C. chloropyga* and *C. albiceps* in spring. There was no delay in oviposition except in winter where a five day delay occurred on the wrapped carcasses. Extremely high internal temperatures in the warmer seasons caused significant maggot mortality on the wrapped carcasses. The predation of *C. marginalis* maggots by *C. albiceps* maggots was observed in autumn. In summer, maggot masses of these species were observed in separate groups. The coleopteran community was dominated by *Thanathopilus micans* L. larvae, succeeded by *Dermestes maculatus* De Geer, larvae. Adult *D. maculatus* and *Necrobia rufipes* De Geer, were present on the carcasses throughout all seasons.

Ectoparasites and arthropod-borne parasites of brown rat, *Rattus norvegicus*, in Ahvaz, southwest Iran

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Arthropods play an important role in the transmission of infectious agents to human and animals, such as rodents. Ahvaz, southwest Iran, is encountered with high density of rodents, especially *R. norvegicus*. In this study, 72 *R.norvegicus* were captured from different sites of Ahvaz, using live traps. Their ectoparasites were collected and identified. Thin and thick blood smears were prepared. After dissection of rats, their different organs were examined for any endoparasites, applying specific techniques. In general ectoparasites comprised 2 species of flea, *Xenopsylla astia* and *X. nubica*, 1 species of louse, *Polyplax spinula* and 1 species of mite *Laelaps nuttalli*. Among endoparasites, 9 species utilize arthropods as vector or intermediate host in their indirect life cycles. These included 1 species of protozoan parasite, *Trypanosoma lewisi* and 8 species of helminth parasites. The potential risk of each of these identified species to the health of human and animals will be discussed.

Applications of GIS and remote sensing for surveillance and control of mosquito borne diseases

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The risk of transmission of mosquito borne diseases depends on the spatial distribution of vector species in relation to the distribution of human hosts and, in case of mosquito-borne zoonoses, the distribution of vertebrate reservoir hosts. These distributions and temporal changes in these spatial patterns depend on environmental and climate/weather conditions. Satellite imagery provides means to gather vast amount of landscape/landcover and meteorological data that are proximate determinants of vector distribution and risk of disease transmission. Geographical information systems allow for the integration of environmental, entomological, epidemiological and demographic data and provide a framework for surveillance and control on scales ranging from local health departments and mosquito control districts to national and international programs. Products of applications of these tools and quantitative analysis of spatial data include data management tools, disease risk maps, and decision making tools for location and time of interventions. Examples from studies in the U.S. Israel, Trinidad and Kenya will be presented, including malaria cases in relation to *Anopheles* larval sites in Trinidad and Israel, dengue and dengue haemorrhagic fever in Trinidad, eastern equine encephalitis outbreaks in Michigan, USA, and La Crosse encephalitis transmission and the effect of introduction of *Aedes albopictus* in Illinois, USA.

Suppression of dengue transmission by application of integrated vector control measures at sero-positive GIS-based foci: Demonstration of a cost-effective model

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A serological survey of approximately 2,000 primary school children from six schools in Chachoengsao, Thailand, was performed at the end of the peak dengue transmission period. GIS analysis of sero-positive cases was carried out to determine transmission foci in high IgG-IgM positive villages. Vector control implementation was conducted in the foci and also within 100 meters around the foci, including schools attended by children in the treated areas, with collaboration from local public health and school authorities. Vector control measures included source reduction by routine garbage pickup organized by local government, the use of locally made screen covers, a combination of local Bti and *Mesocyclops*, and locally made lethal ovitraps. Implementation of vector control measures in the foci was continued until the end of the rainy season. Vector control efficiency was monitored using both entomological and serological parameters. Results showed a significant reduction of dengue vectors in treated areas when compared to untreated areas. Despite overall higher dengue incidences in the second year when compared to the first year, IgG-IgM positive students in the treated areas decreased from 13.46% to 0% while those from untreated areas increased from 9.43% to 19.15%.

Novel Mosquito Traps: Do They Really Work

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A diversity of commercial traps for mosquito control have become available during the past five years. Most are marketed to homeowners. Many models have been evaluated in a large outdoor screened enclosure against two species (*Aedes aegypti* and *Ochlerotatus taeniorhynchus*) of laboratory reared mosquitoes to determine their relative efficacy. Several models were selected for field testing against natural populations. Ongoing research is trying to answer the question: Can mosquitoes be trapped out? Different management strategies are being evaluated, e.g. control potential on isolated islands versus urban/suburban residential settings. In urban/residential settings traps are being evaluated as a protective barrier around a city block and in individual yards. Preliminary results indicate that protective trap barriers provide excellent control (>90%) on isolated islands, but only moderate (50% reduction) control in urban/suburban areas. A single trap in a single yard provided no measurable mosquito control.

The mode of action of deet, SS220, and Bayrepel® arthropod repellents against *Aedes aegypti*, *Anopheles stephensi*, and *Phlebotomus papatasi*

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Laboratory bioassays were conducted with human subjects using six-celled K&D modules and the known arthropod repellents, N,N-diethyl-3-methylbenzamide (Deet), (1S, 2′S)-methylpiperidinyl-3-cyclohexene-1-carboxamide (SS220) and 2-(2-hydroxyethyl)-1-piperidine carboxylic acid 1-methylpropyl ester (Bayrepel) against *Aedes aegypti* (Linnaeus), *Anopheles stephensi* Liston mosquitoes and the sand fly, *Phlebotomus papatasi* which are vectors of human diseases. When five females of each species were simultaneously exposed to one half repellent-treated (4.8×10^{-2}μmol/cm^2) and one half untreated skin beneath the opening of a 3x4 cm cell, they bit exclusively on untreated skin. When treated and untreated skin was covered with cloth to preclude tactile contact with skin surfaces, the insects bit only through cloth covering untreated skin. The tests proved that each species was exquisitely capable of avoiding the compounds in their close-range feeding behavior and this was accomplished by olfactory chemical sensing. When cloth, untreated and repellent treated, was placed over the skin all species avoided the compounds and fed through the cloth only where repellent was absent. In the presence of any the three repellents within a cell, biting responses were reduced by about 50% relative to controls. Thus, besides inhibiting feeding on repellent-treated skin, the compounds also exhibited spatial-repellent activity.

Key issues in advancing the sterile insect technique for african malaria vectors

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Pilot projects that assessed the feasibility of the sterile insect technique (SIT) against Anopheles mosquitoes were conducted in El Salvador in the 1970s. Though on a relatively small scale, these trials demonstrated that sterility could be induced in a wild mosquito population through releasing (chemo) sterilized males. Since then, major scientific advances merit a renewed look at SIT and its applicability in the African context. We describe the R&D phase currently underway in Sudan and Austria, to develop appropriate mass-rearing systems and sterilization procedures. The salient features of such releases, also in relation to the use of transgenic approaches, will be discussed. These include colonization and mass rearing, (genetic) sexing, mosquito fitness and release mechanisms.

The influence of burning on the decomposition and insect succession of pig carcasses

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Recent casework raised the question whether burning has an influence on the decomposition and insect succession of bodies. This was investigated by dousing three pig carcasses with different volumes of petrol and burning them to different degrees (Slight = 1.5L Medium =5L, Heavily = 10L). The insect succession onto these carcasses was compared to a control carcass. The control carcass decomposed at the fastest rate with the heavily burnt carcass at the slowest rate. Adult insects comprised the following: At the control carcass Diptera (60.8%), Coleoptera (29.9%) and Hymenoptera (8.8%). At the slightly burnt carcass Coleoptera (55.8%), Diptera (38.6%) and Hymenoptera (5.4%). The dominant orders at the medium burnt carcass were Coleoptera (38.5%), Diptera (33.3%) and Hymenoptera (28%). At the heavily burnt carcass Coleoptera (51.4%) and Diptera (28.6%) were dominant. Hymenoptera (18.7%) did not have a direct influence on decomposition, since they functioned as predators. *Chrysomya albiceps* was the dominant Calliphoridae species at the control and medium burnt carcasses and *Chrysomya marginalis* at the slightly and heavily burnt carcass. Representative samples of larvae collected during decomposition revealed that *C. albiceps* larvae preferred to feed underneath the carcasses, with *C. marginalis* larvae feeding mostly inside and at the top of the carcasses.

The study of excito-repellency phenomenon of *Anopheles stephensi* exposed to nets impregnated with cyfluthrin

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The excito-repellency phenomenon is occurred in the vector population following the application of definite insecticides. Based on the field observation, in relation to residual spraying operation, the excito-repellency phenomenon may led to behaviouristic changes of the mosquitoes as diversion either to new hosts or other unsprayed shelters. This occurrence may lead to failure in control program. At these conditions, the mosquitoes leave the sprayed houses to either unsprayed or outdoor shelters before picking up the lethal dose. The result of this investigation will be useful for planning of the control program in the country. For the first time, the standard design of excito-repellency (ER) test box was made of aluminum. The laboratory test was carried out with cyfluthrin treated nets against *An.stephensi* (Beech strain). The result showed that the tendency of the mosquitoes to long resting on the netting of exposure chamber was decreased at high dosage of insecticide, and tried to leave the exposure chamber toward the exit trap. The mean of the feeding rate was least at higher dosage in comparison of lower one.

Human behaviors and environmental factors elevating the risk of exposure to *Ixodes pacificus* nymphs in a hardwood forest

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The nymph of the western black-legged tick, *Ixodes pacificus*, has been implicated as the primary vector of the Lyme disease spirochete, *Borrelia burgdorferi*, in northwestern California. In 2002 and 2003, six human behaviors and environmental factors were evaluated as potential risk factors for acquiring nymphal ticks in a dense woodland in this region. The prevalence of *B. burgdorferi* and the causative agents of human anaplasmosis (*Anaplasma phagocytophilum*) and human monocytic ehrlichiosis (*Ehrlichia chaffeensis*) also were determined in nymphs that had been collected from subjects or by dragging leaf litter. Time-adjusted tick-acquisition rates demonstrated that sitting on logs was the riskiest behavior followed, in descending rank, by gathering wood, sitting against trees, walking, stirring and sitting on leaf litter, and sitting on litter. The number of ticks acquired appeared to be unrelated to the type of footwear worn. Nymphal ticks were active throughout the daytime over a wide range of micrometeorologic conditions. Overall, about 3-4% of nymphs were infected with *A. phagocytophilum* and *B. burgdorferi*, regardless of the collection method, versus none with *E. chaffeensis*. Co-infection with *B. burgdorferi* and *A. phagocytophilum* was detected for the first time in this life stage of *I. pacificus*. The public health implications of these findings will be discussed briefly.

Genetically Modified Mosquitoes: A Real World View

G.C. Lanzaro

The concept of controlling mosquito-borne diseases via the development and implementation of genetically modified strains has been under serious consideration since the early 1990's. Although this idea has been met with great enthusiasm in some quarters there remains substantial skepticism in others. The general concept and progress toward the development of such a strategy will be presented. Numerous roadblocks to the development of a GM mosquito plan remain, including (i) lack of gene drivers, (ii) lack of robust effector molecules and mechanisms to manipulate their expression, (iii) poor efficiency of transgenesis, (iv), and (v) no consideration of logistical problems associated with implementation in the field. These will be presented and prospects for overcoming them discussed. Aspects of the ecology and population genetics of targeted species will be discussed as they relate to the successful release and establishment of GM mosquitoes in nature.

Reverse chemical ecology: chemical prospecting of mosquito attractants and repellents

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For a few decades, chemical ecologists have been utilizing insect antennae as the sensing element or biodetector, in electroantennogram (EAG) and gas chromatography-electroantennographic detection (GC-EAD) assays for the identification of minute amounts of pheromones and other semiochemicals. The "molecular" equivalent of these electrophysiology-based "short cut bioassays" is the screening of attractants and repellents based on their binding affinities to odorant-binding proteins (OBPs). OBPs assist hydrophobic semiochemicals to cross an aqueous barrier in the insect sensilla (sensillar lymph) and reach odorant receptors (ORs). Both OBPs and ORs contribute to the high specificity of the insect olfactory system by forming two layers of filters. Although binding per se does not necessarily imply a physiological function, test compounds that do not bind to OBPs can be eliminated from further behavioral studies as they will never "see" the receptors. Reverse chemical ecology is a protein-based approach that requires the full identification of OBPs, cDNA cloning, functional expression of OBPs, and binding assays. In this presentation, we will discuss the latest development of a NIH-funded project* aimed at the development of better attractants and repellents for *Culex* species, the principal vectors of West Nile Virus.* NIH-National Institute of Allergy and Infectious Diseases (1U01AI058267-01).

Developing methodologies for a fungal biopesticide against *Boophilus* microplus, the cattle tick

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The Queensland department of primary industries and fisheries has been investigating fungal biopesticides for livestock ectoparasites as part of a wider strategy to reduce the reliance upon chemicals in livestock industries. Fungal biopesticides have been available for the control of agricultural and horticultural pests for over thirty years and methodologies and accepted protocols for the development of these fungal biopesticides are now well established. Current work with the cattle tick has shown that livestock pests present additional challenges, some of which are due to the inability to conduct *in vitro* assays on obligate parasites, the cost of *in vivo* experiments using animals and the different environments the fungus must operate in when invading the parasitic and free-living phases. In addition, the development of formulation and application methods suitable for penetrating an animal's hair coat and delivering a lethal dose of spores to ticks present further challenges. Solutions to these problems have sometimes been obvious and straightforward but mostly have been arrived at slowly and painstakingly. We have concluded that the development of methodologies for fungal biopesticides of livestock ectoparasites is a multidisciplinary exercise requiring a thoughtful balance of appropriate *in vitro* bioassays with on-animal investigations.

Increased densities of vector mosquitoes in anthropogenic environments in the Kapiti region in New Zealand and potential ecological mechanisms as to why

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Landuse changes, including deforestation, agriculture and urbanisation, have coincided with increases in mosquito-borne diseases worldwide. Anthropogenic environmental changes can alter mosquito populations by modifying the characteristics of breeding habitats, but we still poorly understand the ecological mechanisms involved. Mosquito breeding habitats were sampled in native forest, pastureland and urbanland in the Kapiti region, New Zealand, and 17 environmental characteristics were measured of each. Higher immature mosquito densities were collected from mainly artificial containers in urbanland and pastureland compared to mainly ground pools in native forest. Nine environmental variables were significantly different between landuses but none of these were significantly related to mosquito density. Principal components analysis partitioned environmental data into seven ecological types. One ecological type was significantly related to both landuse and mosquito density, and positively associated with ammonia, bacteria and dissolved organic carbon (DOC), and negatively associated with nitrate and predatory damselfly larvae. The findings of this study support those from prior surveys in warmer climes of greater vector disease risk in anthropogenically-modified environments because of compromised ecosystem integrity. Unlike most previous field-based work, this study also suggests important mechanisms involved; pinpointing mainly bacteria, DOC, nitrate and damselfly larvae as acting in combination to regulate vector mosquito density.

The final word on the genetic, evolutionary and taxonomic relationship of *Pediculus humanus* and *P. capitis*

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Body lice are thought to have diverged from head lice when humans began to wear clothes. This recent evolutionary divergence and incomplete lineage sorting has complicated assessments of whether head lice and body lice are separate species. We used microsatellite markers to test lice from hosts that were infested with head and body lice simultaneously, to see if the head and body lice had interbred. Our results showed that head and body lice had not interbred on a host. Assignment tests revealed that lice that had migrated among hosts always moved from head to head, and body to body, but never between heads and bodies. We conclude that head and body lice are separate species. We speculate that head lice are descended from the lice of neanderthals and modern humans, and that body lice are descended from the lice of modern humans that left Africa about 100,000 ya.

The search for natural repellents from human beings against mosquitoes

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Blood sucking insects are differentially attracted to human beings. This is likely to be caused by differences in the semiochemical profiles of individual human hosts. Despite substantial research into insect-human interactions, the responsible semiochemicals have not yet been identified. An explanation for this difference is that unattractive individuals either lack attractant chemical compounds or may produce compounds that 'mask' the activity of attractants. A special air entrainment technique was developed and used to collect volatile chemicals from human volunteers, and the extracts analysed by gas chromatography (GC), coupled GC-electroantennography (GC-EAG) and GC-mass spectrometry (GC-MS). For the first time electrophysiological responses of female yellow fever mosquitoes, *Aedes aegypti* to volatiles from air entrainment extracts were recorded, and a number of electrophysiologically active compounds were identified. Previous behavioural studies have confirmed the differential attractiveness of volunteers to biting insects. A quantitative and qualitative analysis of compounds within the extracts, from individuals known to be differentially attractive to biting insects, has revealed significant differences in chemical profiles. The identification of behaviourally active compounds could lead to new and improved control technologies, whereby 'masking' compounds could potentially be incorporated into new, safe and natural repellents against biting insect pests.

Anopheles arabiensis at the edge of its distribution in northern state, Sudan

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Northern state, Sudan is mainly desert, rainfall is rare and most life is found close to the Nile. *Anopheles arabiensis* is the only anopheline and malaria vector found in the area in recent decades. It is present along the Nile downriver to about 200 km south of the Egyptian border, where poor terrain and control efforts prevent its spread further north. Initial results are presented from field and molecular studies to determine the population structure and dynamics of the species and related environmental factors. The receding Nile following the flood contributes most to sustaining *A. arabiensis*. Larval breeding sites can be found on low lying riverbank, ied-up inlets, or channels around islands, through most of the year. Land use is mainly associated with irrigation, but this does not appear to contribute greatly to anopheline numbers, however the extent of human presence and activities do increase the potential for anopheline breeding. Upriver from the fourth cataract and downriver from the third cataract, the terrain is rocky desert and less suitable for both humans and mosquitoes. The *An. arabiensis* population is thus semi-isolated within an approximately 700 km corridor a few kilometres wide and breaks down further into pockets depending on particular river features.

Control of biting midge (Ceratopogonidae) attack on residents of a marine canal estate in Redcliffe (Australia) by residual barrier treatment of Bistar 80SC® to ornamental garden plants in house yards

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Biting midge (Ceratopogonidae) breeding in marine sediments associated with engineered canals in a residential community in Redcliffe Australia have been responsible for considerable biting attack and nuisance. Limitations on approved applications of larvicides in breeding sites has historically restricted control to personal protection, midge screening and education of residents. In October 2003 – Feb 04, Redcliffe City Council funded a trial to test the efficacy of Bistar 80SC® (FMC Chemicals Pty Ltd) as a 0.1% AI (Bifenthrin) residual application to ornamental garden vegetation and dwelling external walls. Eight paired randomised treated and untreated premises were assessed for differences in biting midge exposure. The assessment was undertaken using CO_2 and Octenol baited light traps placed in treated and untreated yards. A self administered questionnaire scoring individual household daily experience of biting midge activity was also used. Untreated control premises received a placebo application of water alone to control bias in the resident questionnaire. Analysis of difference in light trap collections between treated and untreated showed a 76.4% reduction in midge (primarily *Culicoides molestus*) from treated premises. Resident survey results indicated occupants of treated premises experienced generally favourable biting midge conditions while residents from untreated homes experienced fluctuating exposure and discomfort corresponding to lunar emergence cycles.

Aerial efficacy assessment of Anvil 10+10 against *Anopheles quaimaculatus*

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Aerial ULV applications of Anvil 10+10 were conducted against caged, field-collected *Anopheles quaimaculatus* adults using Beecomist® 360A rotary atomizers. Standard and optimized applications were conducted at 0.62 fl oz (0.0036 lbs a.i.) per acre using stacked passes 3,500 ft and 5,000 ft upwind of the target site. Efficacy of Anvil 10+10 and impact of wind velocity at flight altitude (150 ft) were assessed by bioassay and analysis of collected droplets.

The potential role of aquatic insects in the transmission of buruli ulcer disease in Africa

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Buruli ulcer is the third most frequent mycobacterial disease in humans, next to tuberculosis and leprosy, often causing serious skin ulcerations, bone deformities, and disability. Unlike TB and leprosy, Buruli ulcer has emerged dramatically since the 1980's, with increasing numbers of cases being reported from at least 31 countries, mostly in tropical and subtropical areas, including Africa. Although poorly understood, the epidemiology of Buruli ulcer associates the resurgence with people who live and/or work close to wetlands, especially slow flowing (riverine) or stagnant water bodies (marshes, swamps), often created as a result of some human environmental disturbance. The disease is not known to be contagious between humans, and epidemiological studies have not yet established natural reservoirs or modes of transmission. However, it was recently demonstrated experimentally that *M. ulcerans* could survive and multiply exclusively within salivary glands of aquatic bugs (Naucoridae: *Naucoris* sp.) and they were able to transmit *M. ulcerans* to mice by their bites, suggesting a possible mechanism for human infection. The organism has been found in samples of water, biofilm and detritus. A possible trophic scenario for the natural mode of transmission of *M. ulcerans* is presented based on preliminary work in Ghana by the authors.

Efficacy of a Spinosad Suspension Concentrate Formulation for Controlling Chewing and Sucking Lice on Cattle Under Field Conditions

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Field studies were conducted with cattle in Wisconsin and Illinois between January and April, 2000, to determine the efficacy of 0.04% spray and 2 mg/kg pour-on treatments of a spinosad suspension concentrate formulation against chewing and sucking lice. Positive control substances included in the study were Co-Ral® EC (5.8% coumaphos) and CyLence™ Pour-on (1% cyfluthrin). Animals in the negative control group remained untreated. All lice species were visually sampled on each animal at weekly intervals after treatment, for eight weeks. Individual louse counts were summed over each examination site for each species. A repeated measures analysis using a mixed model (SAS PROC MIXED), was conducted for data pooled across both study sites. Both spinosad treatments and CyLence™ Pour-on provided excellent efficacy (98-100%) against *Bovicola bovis* over the entire study period; Co-Ral® EC efficacy opped below 90% from Weeks 6-8. The efficacy of the spinosad spray treatment against *Linognathus vituli* was >93% for the entire study and >88% for the spinosad pour-on treatment. Co-Ral® EC was >90% efficacious against *L. vituli* during Weeks 1-7, while CyLence™ Pour-on only exceeded 90% for Weeks 3-7. *Solenopotes capillatus* were found only at the Wisconsin site; all treatments were highly efficacious throughout the study period.

The effect of human age on dengue seroconversion risk in Iquitos, Peru

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Basic models of infectious disease often assume that force of infection is independent of host age. This assumption has been shown to be violated for many directly transmitted diseases, but has rarely been examined in vector-borne systems, such as dengue. Data from DNA fingerprinting studies of mosquito bloodmeals indicate that the biting rate of *Aedes aegypti* increases with human age. If risk for dengue infection increases with human age, models assuming an age-independent risk will underestimate the proportion of the population that needs to be vaccinated in order to eliminate disease transmission. A cohort of approximately 3,000 schoolchildren and their families in Iquitos, Peru were followed for up to 5 years (1999-2003) with blood samples collected at 6-month intervals. Sera were tested for dengue virus antibodies by the plaque reduction neutralization test. The effect of human age on risk for dengue seroconversion was evaluated using a multiple logistic regression model, which also included area of residence within the city. Preliminary analysis suggests that risk increased with age and was highest in the southern region of the city. Estimates of age-specific forces of infection can be used to design an optimal immunization program.

Oviposition Site Selection by Mosquitoes in Response to drought

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Mosquitoes are vectors of many important medical and veterinary diseases. Although outbreaks of mosquito borne disease can be correlated to changes in precipitation in the very short term, precipitation rarely corresponds to mosquito abundance in the long term. Here, I present the results of a field based mesocosm experiment designed to test how water level and drought influence the oviposition behaviour of two species of mosquitoes. I used two water levels (high and low) and two drought treatments (normal and drought). Water for the drought treatments was obtained from ponds that had experienced ying for a period of a month prior to experimentation. The two mosquito species differed markedly in their oviposition response. The egg laying behavior of *Ochlerlatatus notoscriptus* was significantly greater in the drought treatments regardless of water level. However, the egg laying behavior *Anopheles annulipes* showed the opposite response with significantly more larvae present in the high water treatments regardless of whether the treatment water was normal or in drought. My results provide supportive evidence that mosquitoes may show population outbreaks following drought years and that this framework may provide a reliable way of predicting year-to–year variation in mosquito abundances based on individual species oviposition response.

Tests on a new bait for strike flies (Diptera: Calliphoridae)

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A bait consisting of sulphur-containing volatile compounds encapsulated in a caesin-based slow release matrix was tested for its efficacy in catching sheep strike flies (Diptera, Calliphoridae). The fresh bait was significantly more effective than a liver/sodium sulphide mixture in trapping a major strike fly (*Lucilia sericata*) and a minor strike fly (*Chrysomya rufifacies*) on a sticky trap. The bait also trapped significantly more gravid flies of these species. The bait efficacy wore off after 17 days under ambient conditions. The bait was also less effective than the liver mixture when used in Western Australia traps. This may be due either to its loss of effectiveness with age, or to differences in the responses of flies when landing close to the bait on a sticky trap and when searching for the exact location of an odour source in a Western Australia trap. Ways of improving the bait efficacy are discussed.

Aedes aegypti surveillance in Iquitos, Peru: a 4- year test of the pupal/demographic survey

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We present results from 4 years of pupal/demographic surveys of a circuit of ≈ 6,000 houses surveyed >10 times in the Amazonian city of Iquitos, Peru. We compare traditional larval indices with pupal and adult mosquito indices. We evaluated *Aedes aegypti* production from different container classes, including those based on water management practices overtime and space. A total of 289,941 water-holding containers were characterized, of which 7.3% were positive for *Ae. aegypti*. Although, temporal and geographic variations were detected for all variables examined, consistent production patterns were observed for the lid status (lids: 32% wet containers, 2% pupal production), container location (outdoor: 43% wet containers; 85% pupal production) and method by which the container was filled with water (rain filled: 15% wet containers; 88.3% pupal production). Based on these patterns and additional targeting of containers with highly variable pupal counts we propose a targeted control strategy for Iquitos that would reduce *Ae. aegypti* populations below published but untested threshold values for Iquitos.

Screening medicinal plant extracts for effects on *Aedes aegypti* (Diptera : Culicidae) and toxicity to a non target organism

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A preliminary study was conducted to find out the effects of the extracts of 112 medicinal plant species, collected from the southern part of Thailand, on *Aedes aegypti*. Studies on larvicidal properties of plant extracts against the third and fourth instar larvae of *Ae. aegypti* showed fourteen species with high toxicity. Six plant species were comparatively more effective against the larvae at very low concentration. These extracts demonstrated no or very low toxicity to guppy fish (*Poecilia reticulata*). Three plants with promising larvicidal activity, *Mammea siamensis*, *Anethum graveolens* and *Annona muricata,* were used to study of the impact of the extracts on the life cycle of *Ae. aegypti*. The results showed that these plants affected the reproductive potential of surviving adults by reducing the number of eggs laid and egg hatchability. The first instar larvae were very susceptible to *A. muricata* and the second instar larvae were susceptible to *A. graveolens* while the third and fourth instar larvae were susceptible to *M. siamensis*. These extracts delayed larval development and inhibited adult emergence.

The mosquito burden in Kenya

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Kenya is a tropical country with a climate favourable for diverse insect life – both the harmful and the useful alike. Embu District experienced a malaria outbreak between January and February 2001 where more than 300 people were admitted at the main health facility in the District, 70 lives were lost. Malaria is endemic in Embu district with a morbidity of 60% and mortality of 25%. Expectant mothers and children are affected most with slide positivity of 30% and 45% respectively. Annually, each household spends around US$ 65 on malaria control and funeral expenses. Larval densities range from 30-40% with peak densities in March, April and May. Despite this, 25% of the community cannot positively identify mosquito as the malaria vector. Only 12% can correctly name the five main symptoms of malaria (fever, chill, sweating, anorexia, joint pains). The ministry of health has issued guidelines on uniform treatment and community sensitisation. Consistent use of bednets, chemoprophylaxis and indoor residual spraying are being encouraged. Plans are underway to start biological adult and larval control. This paper discusses the mosquito burden from the perspectives of larval density, host prevalence index, mortalities and morbidities, bed occupancy and the economics of vector control

Tick vector-pathogen interrelationships

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The interrelationships between tick-borne pathogens, their tick vectors and diverse vertebrate hosts, are represented by a triangle of parasitic interactions. The interactions are between (i) tick-pathogen, (ii) pathogen-host, and (iii) tick-host. In (i) the pathogen interacts with its vector, infecting and replicating within tick cells or extracellular spaces. The ability of a particular tick species to act as a vector depends on whether the pathogen can survive and overcome several 'barriers' within the tick. However, this is a simplistic view of the tick vector-pathogen interrelationship as both (ii) and (iii) influence the success of (i). This triangle of interactions is focused at the skin site of tick feeding - the initial site at which pathogens gain access to either their host or vector. For pathogens, the skin-feeding site is an ecologically privileged niche, profoundly modified by the pharmacological activities of bioactive tick saliva molecules. Exploitation of this niche is known as saliva-activated transmission (SAT). Tick saliva molecules that promote SAT have not been identified but appear to differ for different tick vector-pathogen interrelationships. Identifying the SAT factors and their mode of action offers new insights for controlling ticks and tick-borne diseases.

Argentina: veterinary forensic entomology as a potential tool against rustling

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Cattle rustling entails economic loss to cattle owners and danger to their safety, marketing of uninspected meat and hides and loss to the State from unpaid taxes. In 2003, the Faculty of Veterinary Medicine (Tandil, Buenos Aires province, Argentina) incorporated forensic entomology to a course of legal veterinary medicine aimed at fighting rustling. Preliminary field experiments showed a large proportion of hemisynanthropic insects, most of them Neotropical, which may represent the autochthonous necrophagous fauna, now displaced by synanthropic introduced species in urban environments. Colonization and development times proved longer in the arid climate of Tandil than at Buenos Aires city, where most published data were gathered. Field experiments whith bovine bodies and parts are required. Hitherto, bovines have been avoided as a model because of the added complication of the ruminant digestive tract. Since most countries with rustling problems also have a pattern of introduced species displacing the original necrophagous fauna in city environments, they might benefit from knowledge obtained through these studies.

Arboviruses, mosquitoes, and RNA interference

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The identification of critical arbovirus-vector interactions reveals new insights into vector competence. For instance, flavivirus and alphavirus replication in mosquitoes triggers a natural RNA interference (RNAi) response; however, viral counterdefenses probably allow RNAi evasion and establishment of persistent infections. We can enhance RNAi to dengue virus type 2 (DENV2) in *Aedes* species by administration of double-stranded (ds) RNA derived from the DENV2 genome, either by direct transfection of dsRNA, transient expression from a Sindbis virus (SINV) transducing system, or stable transformation with a plasmid that expresses a hairpin-forming transcript. RNAi is sequence-specific, is maintained by low concentrations of effector RNA, blocks accumulation of intracellular viral RNA, and results in the appearance of 21-25 nt siRNA homologous to the mRNA target. We have identified genes from the Dicer and Argonaute families in 2 mosquito genera and have used RNAi to knock down their expression to determine which genes have a role in virus resistance. We have evidence that DENV2 and some strains of SINV establish persistent infections in mosquitoes by suppression of RNAi, and are investigating mechanisms. We are identifying viral-encoded suppressors by site-specific mutations in infectious cDNA clones. Our work suggests that persistent infections in mosquitoes result from a balance between host RNA silencing and viral counterdefenses and provides new strategies for tipping the balance in favor of the vector host.

Endosymbionts and sex ratios in the human head louse

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At the turn of the 19th century the first observations of a female-biased sex ratio in broods and populations of head lice had been reported. A study by Buxton in 1938 on the sex ratio of lice on prisoners in Ceylon is still today the subject of reanalyses. However, in the last sixty years no data have been collected, especially on more normal levels of infestations. Here we report a female bias of head lice in a survey of 480 school children in Argentina. This bias is independent of lice intensity. These lice as well as lice from pupils in Britain were carrying several, related strains of the endosymbiotic bacterium *Wolbachia*, one of the increasing number of intracellular sex ratio distorters. No lice without *Wolbachia* were detected. Similar *Wolbachia* strains are also present in the pig louse suggesting that this endosymbiont might have marked influence on the biology of the whole order. The sex ratio distortion in head lice that has been reported world wide is stable over time; that is a remarkable deviation from the stability of frequency-dependent selection of Fisher's sex ratio.

Blood Feeding in *Cnephia* (Diptera: Simuliidae): Molecular Phylogeny

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Five species of *Cnephia* were studied, three non-biting, one a mammal feeder, and one a bird feeder. Of the non-feeding species, *C. eremite* is Holarctic in distribution and specimens were examined from both Canada and Sweden. *Cnephia pallipes* is a Palearctic non-feeder and *C. dacotensis* a temperate Nearctic non-feeder. *Cnephia pecuarum* is an economically important large mammal feeder in south-central U.S. and *C. ornithophilia* a widely distributed bird feeder in southern and eastern North America. Sequence was obtained from the complete mitochondrial CO-II gene (687 bp) and partial EF1-alpha nuclear gene (1030 bp). Molecular data is discussed in relation to conventional wisdom about relationships among species based on cytological and biological information, geographic distribution and genetic diversity within species. Molecular data strongly support *Cnephia* as a monophyletic clade in the tribe Simuliini. Speciation hypotheses were not fully concordant between molecular and conventional data.

Onchocerciasis in Central America: Ecology & Control

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Detection of O. volvulus larvae in vector populations of black flies is of prime importance in the assessment of the effectiveness of ivermectin control programs. Detection of larvae is attained by dissection of individual flies or by PCR (amplification of an Onchocerca 150 bp repeated sequence, followed by hybridization of the PCR products with a species-specific DNA probe) from pools of flies. The PCR was compared with dissection method to estimate the infection rate (p) and the mean larval load (any larval stage) per parous fly (m) in a local vector population. Both methods produced statistically similar estimates, thus PCR is an alternative and efficient method for entomological surveillance. Sample sizes required to estimate m from p for fixed levels of precision were calculated using Mexican and Guatemalan data, assuming that k (an aggregation parameter from the larval distribution among parous flies) is known. Optimal sampling and PCR was used to evaluate the impact of ivermectin on m, p and transmission potentials in Mexico. The data suggest that substantial progress towards the goal of suppressing transmission has been accomplished. In southern Chiapas, entomological estimates decreased in comparison with the baseline data, but PCR positivity was still detectable. In northern Chiapas and Oaxaca, with no baseline data, there was an absence or near absence of infected and infective flies.

Malaria vectors, *Anopheles hyrcanus* group (Culicidae, Diptera): morphological comparisons and molecular analysis

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Anopheles hyrcanus group consists of at least 30 species, with several species including *Anopheles sinensis, lesteri, pullus*, that are known or potential malaria vectors. The morphological diagnostic features and molecular identities of malaria vector *hyrcanus* group species are determined. Larvae, pupae and adults of selected species were described mophologically, and their ITS2 and DNA sequences from our laboratory and GenBank were compared.

Distribution and bionomics of malaria mosquito vectors, *Anopheles (Anopheles)* in Japan, Korea and other parts of Asia

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Mosquito specimens of *Anopheles (Anopheles)* from collections conducted in Japan (2002-03), Korea (1999-2003), Philippines (2002), HongKong (2001), mainland China (2001-03) were identified using morphological and molecular techniques, and recorded. Larval, pupal and adult rearings of field collected samples were conducted for morphological and molecular analysis. Detailed locations of various collection sites and habitats were characterized for known and potential malaria vectors of different countries. Habitats of vectors include ricefields, irrigated ditches, temporary and permanent pools, ponds, streams, others.

Dengue mosquito vectors, *Aedes* and *Ochlerotatus*: morphological identifications and distribution

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The diagnostic morphological features of mosquito vectors of dengue viruses are compared for various *Aedes (Diceromyia, Stegomyia)* and *Ochlerotatus (Finlaya, Gymnometopa)* species found in four regions of the world, namely Oriental Region, Pacific and Australian Region, Afrotropical Region, and the Americas. Morphological characters are indicated to separate both larvae and adults of the vector species of each region. The distribution and bionomics of *Aedes* and *Ochlerotatus* vector species are included.

New and resurgent arboviral disease threatening humans in Australia

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Australia has a range of indigenous arboviruses and an introduced few that are of public health concern; all are mosquito-borne. Endemic encephalitogenic viruses (Murray Valley Encephalitis, Kunjin) are annually active and responsible for occasional cases in the northwest but, despite widespread vector distribution, only rare activity and no epidemics in the southeast since 1974. The most common arboviruses, Ross River annually infecting many thousands nation-wide causing polyarthritis, and Barmah Forest (a similar syndrome) on the increase and responsible for outbreaks of hundreds regionally, have a spectrum of coastal saline and inland freshwater vectors. The most important exotics are the returning dengue viruses with outbreaks of hundreds confined to Queensland by vector distribution but with virus endemicity and vector spread threatening, and the recently introduced Japanese Encephalitis virus causing only a few deaths in northern Queensland but with virus persistence and its affinity for a local vector threatening. Environmental factors drive arbovirus activity by influencing vector populations, which vary with virus and circumstance, although little is known of the role of reservoir hosts. Tourism and development activities that increase human/vector/virus contact, and climate changes that promote vector and virus proliferation, threaten to exacerbate the disease threat to the public health.

Repellents for integrated tsetse management

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ICIPE is in the forefront in identifying potent repellents, both synthetic analogues of natural tsetse repellents and natural repellent blends from un-preferred animals like waterbuck and zebras. One potent synthetic repellent for savannah tsetse species, developed by molecular optimization studies, acts as an olfactory antagonist of a key phenolic kairomone, which flies use to locate the host for feeding (Saini & Hassanali; patent application). This repellent substantially reduce tsetse challenge (> 80%) and feeding efficiency (>80%) on cattle. A prototype dispenser which is placed around the neck of an animal has been developed. Field trials indicate that the repellent can provide effective protection to cattle but all animals in a herd need to be protected or else significantly higher infection occurs in unprotected animals. Experiments are ongoing to evaluate how repellents can be integrated with other existing tsetse control techniques. A 'push-pull' strategy that deploys repellents (to 'push') and baited traps / targets (to 'pull') to enhance suppression rates is being evaluated. A variant of this may involve the use of a proportion of cattle with 'pour on' insecticides as live baits. A business plan for commercial production and distribution of repellent products is also being developed.

Mitochondrial genome organization of lice is highly variable

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Mitochondrial (mt) genome organization is extremely conserved in mammals, vertebrates and most arthropods, e.g., humans and frogs have the same mt genome organization; so have fruit flies and water fleas. We have sequenced the entire mt genomes of the wallaby louse (Phthiraptera: Amblycera) and the pigeon louse (Ischnocera), and part of the mt genomes of the human louse (Anoplura) and four other amblyceran lice. Unlike most arthropods, the mt genome organization of these lice is highly variable. Lice share only 1-2 gene boundaries with fruit flies. No gene boundaries were shared among lice of the three suborders. Mt genome organization differs even among three genera within the suborder Amblycera. Changes in the mt genome organization of lice include gene translocation, inversion, separation of rRNA genes and loss or multiplication of non-coding regions. In addition to lice, we found mt genome organization was variable or highly variable in other two hemipteroid orders, Psocoptera (psocids) and Thysanoptera (thrips), but is strictly conserved in the order Hemiptera (bugs). We speculate that the changes in the mt genome organization in lice, psocids and thrips may be related to their life-history traits such as small body size, small colony size, and high degree of inbreeding.

Kinetics of *Borrelia burgdorferi* infection in larvae of refractory and competent tick vectors.

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Borrelia burgdorferi is the causative agent of Lyme disease and is transmitted by the tick *Ixodes scapularis*. The ticks, *Amblyomma americanum* and *Dermacentor variabilis*, represent non-competent or semi-competent species for this spirochete. The present work assesses *B. burgdorferi* acquisition by the larvae of these three species of ticks, detailing the dynamics of the spirochete colonization in these refractory species. *B.burgdorferi* counts were assessed by qPCR using primers and probe for the spirochete *flaB* gene. Larvae were fed on infected mice and the spirochete loads were determined after 3 to 4 days of feeding and up to 28 days post-feeding. Analyses of pools of 10 larvae indicate that *A.americanum* is very refractory to *B.burgdorferi*, with no detection of spirochete DNA during or post-feeding. *I.scapularis* supports high loads of spirochetes (10^3-10^4/larvae). In *D.variabilis*, *B.burgdorferi* uptake was reduced. However, these ticks supported spirochete growth up to the fourth day post-infection, when the ratio of spirochetes increased from 1/300 to 1/7 of the counts in *I.scapularis*. After this time point, *D.variabilis* no longer supports spirochete growth, but *B.burgdorferi* DNA was still detected after molting. These results indicate inappropriate conditions for *B.burgdorferi* colonization and uptake, or the presence of anti-*Borrelia* factor(s) in non-permissive tick species.

West Nile virus activity in the USA and regional differences in vectors and transmission dynamics

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In the USA, West Nile virus (WNV) was first detected in New York City in 1999. By the fall of 2003, WNV had spread south and west to 45 states. During 1999–2003, 13,405 human cases of WNV disease were reported. Principal enzootic and epidemic vectors of WNV in the USA are *Culex (Culex)* mosquitoes. In the Northeast and upper Midwest, *Cx. pipiens* L. including form molestus, *Cx. salinarius* Coquillett and *Cx. restuans* Theobald are the most important vectors. In the South, *Cx. quinquefasciatus* Say and *Cx. salinarius* are the most important vectors. In Florida and the southern portions of LA, MS, and AL, *Cx. nigripalpus* Theobald is a potential vector. In the Rocky Mountain and High Plans states, *Cx. tarsalis* Coquillett and *Cx. pipiens* are the primary vectors. In the arid Southwest, *Cx. tarsalis* is the primary vector with *Cx. quinquefasciatus* playing a role in urban areas. Across much of the mid-latitudes, *Cx. pipiens* and *Cx. quinquefasciatus* hybridize and both taxa and hybrids are important vectors. California will experience WNV disease in the future and important vectors are likely to be *Cx. tarsalis* and members of the *Cx. pipiens* complex, with *Cx. stigmatosoma* Dyar and *Cx. erythrothorax* Dyar being focally important. Important avian host vary with regional abundance, however, the house sparrow (*Passer domesticus*) appears important in virus transmission throughout the USA.

Aedes aegypti **genomics**

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"We have initiated a genome project for *Aedes aegypti* that will ultimately lead to complete sequence information. We are sequencing both ends of genomic DNA Bacterial Artificial Chromosome (BAC) clones and random cDNAs representing a variety of tissues, and physically mapping selected BAC clones. BAC clone end-sequencing is completed and the data are available at GenBank. We plan to sequence and analyze both ends of ~80,000 cDNAs derived from normalized libraries produced from 4 specific mRNA sources and one library enriched for full-length sequences. Individual libraries are in various stages of construction, with one competed (*Brugia malayi*-infected) and 29,824 ESTs generated, and sequencing of a second (*Plasmodium gallinaceum*-infected) nearly completed. The third library (females exposed to a dengue virus) has just been completed and we are in the process of constructing libraries from 1) fat body and 2) combined developmental stages. We have incorporated all sequences from our genome project with gene sequences available in GenBank into an *A. aegypti* Gene Index available at TIGR (www.tigr.org/tdb/tgi/aegi/). We have recently (2/13/2004) been notified that NIH-NIAID has selected *A. aegypti* for full genome sequencing. We anticipate this will include an 8X coverage that should provide a high quality genome assembly. "

Measures of *Aedes aegypti* density and the risk of dengue virus transmission.

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In two large, cohort-based prospective studies we are defining entomological risk and characterizing its relationship to human dengue infection. Based on results from more than 5 years of field research in the Amazonian city of Iquitos, Peru we are determining the appropriate scale (spatial and temporal) at which to measure entomological risk and dengue virus transmission. A new research program in Khamphaeng Phet, Thailand is an extension of the Iquitos project and will explicitly link in time and space various measures of entomological risk to human dengue infections

Mitochondrial genome organization of lice is highly variable

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Mitochondrial (mt) genome organization is extremely conserved in mammals, vertebrates and most arthropods, e.g., humans and frogs have the same mt genome organization; so have fruit flies and water fleas. We have sequenced the entire mt genomes of the wallaby louse (Phthiraptera: Amblycera) and the pigeon louse (Ischnocera), and part of the mt genomes of the human louse (Anoplura) and four other amblyceran lice. Unlike most arthropods, the mt genome organization of these lice is highly variable. Lice share only 1-2 gene boundaries with fruit flies. No gene boundaries were shared among lice of the three suborders. Mt genome organization differs even among three genera within the suborder Amblycera. Changes in the mt genome organization of lice include gene translocation, inversion, separation of rRNA genes and loss or multiplication of non-coding regions. In addition to lice, we found mt genome organization was variable or highly variable in other two hemipteroid orders, Psocoptera (psocids) and Thysanoptera (thrips), but is strictly conserved in the order Hemiptera (bugs). We speculate that the changes in the mt genome organization in lice, psocids and thrips may be related to their life-history traits such as small body size, small colony size, and high degree of inbreeding.

Simuliid taxonomy in relation to transmission of human onchocerciasis and mansonelliasis in Brazil.

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Mansonelliasis was recorded as widespread in the southern Amazon over 50 years ago and human onchocerciasis 35 years ago in a discrete area in the northern Amazon region of Brazil. Incrimination of the simuliid vectors that transmit the filarial parasites responsible for these diseases has only been possible during the last three decades, largely due to the logistical difficulty in collecting species in these regions and the complex taxonomy of members of the *Simulium amazonicum* group to which several belong. This has been caused by the morphological similarity between vector and non vector species of the group. The use of digital image analysis has facilitated the morphological definition of species in the group, recently augmented by cytological and molecular studies. Our current classification of the group based on these techniques is presented together with distribution maps of vector species that suggest that disease dispersal to non endemic areas may be occurring as infected individuals migrate from the disease foci as rural areas of the Amazon are developed.

Polymerase chain reaction-based differentiation of members of the *Anopheles culicifacies* complex

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Anopheles culicifacies, the principal vector of malaria in India, is responsible for over 65% of total malaria in India and is a complex of 5 cryptic species designated as species A, B, C, D, and E. These members are morphologically indistinguishable at any stage of life and can be identified by examining species-specific inversions in polytene chromosome and mitotic karyotype of Y-chromosome of male progeny. In an attempt to develop a simple alternative to cyto-taxonomy, we have developed an Allele-Specific Polymerase Chain Reaction (ASPCR) assay targeted to D3 domain of 28S ribosomal DNA, which unambiguously differentiates *An. culicifacies* species A and D from species B, C, and E. The assay was evaluated using chromosomally-identified specimens of *An. culicifacies* originating from different geographical regions of India representing different sympatric associations and was found to be specific. The culicifacies-specific primers used in the assay did not cross-react to other anophelines as tested against all members of *An. fluviatilis* complex (species S, T, and U), *An. minimus* (species A), *An. stephensi* and *An. subpictus*. The search of genomic database using culicifacies-specific primers sequences revealed that these primers are specific to *An. culicifacies* only. The possible use of this diagnostic assay in disease vector control programme for discrimination of vector species from non-vector will be discussed.

Invasive species impacts, bee and wasp injuries in New Zealand

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The continued global modification of the environment by human activities has resulted in broad scale ecological disruptions, including the introduction of exotic invasive species. In New Zealand over the last two centuries, two exotic species of bees and four exotic species of wasps of public health concern have become established. In 1990 it was reported that NZ had the highest recorded densities of wasps in the world. In order to establish the prevalence of sting injuries in NZ, we analysed NZ health statistics data and carried out retrospective questionnaire based population surveys. The incidence of bee and wasp sting related hospitalisations remained similar over the twelve-year period from 1988-1999, with an average of 3.1/100,000/year, higher than that reported for the USA but lower than Australian hospitalisation rates. While, results from the questionnaires indicated a high prevalence of sting injuries 1,300/100,000/month (based on a six-month period), significantly higher than that reported in other countries. We estimate that the degree of under-reporting in NZ health statistics from sting injuries could be as great as 53-fold. New Zealand's unique environment (leading to high densities of wasps) and New Zealander's outdoor lifestyle may explain the high prevalence of sting injuries.

Insecticides don’t kill lice but formulations of insecticides do!

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Head lice treatment preparations are labelled according to the qualitative and quantitative content of their active ingredients. A recent review of this group of products by the Medicines Evaluation Committee of Australian Therapeutic Goods Administration concluded that "There is a paucity of good quality information on the effects of different formulations on the efficacy of head lice products". This presentation will review the available evidence and identify some of the issues which may impact on dosage form design of head lice preparations.

Potency and avidity of tick paralysis antitoxin in Australia

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Tick paralysis due to the Australian paralysis tick, *Ixodes holocyclus* is a potentially-lethal syndrome affecting domestic pets, livestock and humans along the east coast of Australia mainly during late spring and early summer. There is no vaccine available at present. Prevention in animals largely depends on acaricidal treatment. Treatment of paralysis is based on a commercially-available antiserum, raised in hyperimmune dogs by progressive and cumulative antibody stimulation by repeated infestation with adult female ticks. This serum constitutes a specific, antitoxin widely used by veterinarians and to a limited extent by physicians to treat cases of tick paralysis. Standardisation at the minimum arbitrary level of 500 antitoxin units per ml is legally required. Variability occurs in potency and particularly in avidity (the well-known relative variability on dilution to neutralise an active toxin [in this case, holocyclotoxin] *in vivo* and *in vitro*). A modified method using dilution of antitoxin as opposed to the earlier method of dilution of toxin, may be used to study this variability. This variability will be discussed and possible explanations proposed.

Global expression profiling of the major detoxification enzyme families in the major African malaria vector *Anopheles gambiae*

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Vector control programs based on the use of insecticides, whether it be by residual house spraying or treated bed nets, are being challenged by the emergence of insecticide resistance. For example, there is wide scale resistance to pyrethroids in mosquitoes from West Africa, which are the major insecticides used in malaria control. One reason for this is the increased detoxification of insecticides by three major enzyme families; glutathione *S*-transferases, cytochrome P450 monooxygenases and the carboxylesterases. The recent publication of the complete genome sequence of the major malaria vector *Anopheles gambiae,* has allowed us to construct a highly specific 'detoxification chip', which contains fragments of all the genes putatively implicated in insecticide resistance in this mosquito. We have used microarray technology to monitor the expression of genes in insecticide-susceptible and resistant mosquito strains to pin-point the genes responsible for resistance. Furthermore, to generate a more detailed profile, their expression has also been assessed in the various life stages of the mosquito and in male and female insects. The detoxification chip is an excellent tool to provide a platform with which to go on and determine the function of these enzymes.

Odour blends in the host-seeking behaviour of the malaria vector *Anopheles gambiae*.

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The mosquito *Anopheles gambiae* is the most important malaria vector in Africa primarily because of its strong anthropophilic behaviour. We have studied factors affecting host-seeking behaviour of this mosquito, notably the olfactory cues with which human hosts are being detected. Responses of the mosquitoes to human skin emanations were studied with electrophysiological and behavioural essays. We demonstrated that important olfactory stimuli for *An. gambiae* are ammonia, L-lactic acid and several aliphatic carboxylic acids. EAG and single sensillum recordings demonstrated significant dose-responses in olfactory cells of grooved peg and trichoid sensilla. Behavioural responses were observed with single and mixed odours in an olfactometer. *An. gambiae* always responded to ammonium, although as a single component ammonium could not compete with the natural odours from human skin. Ammonia was an essential synergist to cause activity of lactic acid and/or carboxylic acids. Laboratory results were complemented by field studies in West Africa, where it was found that wild *An. gambiae* were attracted to a synthetic blend of volatiles from a distance. The same blend of odours that caused high attraction in the olfactometer, attracted most mosquitoes to odour-baited traps. The results are discussed with consideration for the development of olfaction-based technologies for malaria control.

Novel mosquito repellents derived from essential oils of plants in Thailand

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Mosquito-borne diseases, such as dengue, malaria and filariasis are serious public health problems in many countries, especially in the tropical regions of the world. These diseases are caused by pathogens that are transmitted to man through mosquito biting only, thus personal protection from mosquito bites is then considered as one of the strategies to prevent them. Efforts to develop alternatives to replace chemical repellents have been increasingly considered, including extracting and evaluating plant products for repellent activity. The development and use of locally available plant products showing repellent activity thus avails an alternative strategy for prevention of mosquito-borne diseases. In this study we evaluated and reported repellent effects of essential oils extracted from plants against 4 mosquito vectors: *Aedes aegypti*, *Ae. albopictus*, *Anopheles dirus* and *Culex quinquefasciatus* under laboratory conditions using human volunteers. The essential oils were extracted from various parts of 22 plant species, belonging to 10 families, and the oils were prepared as 10% solution in absolute ethanol and some additives. The essential oils showing high degree of repellency were subsequently formulated as topical repellents and tested against day- and night-biting mosquitoes under field conditions. The results obtained from both laboratory and field evaluations will be presented.

Evaluation and development of cockroach repellents derived from essential oils of plants

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Cockroaches are common insects found throughout the world, especially in the tropical regions. They are generally recognized as not only household pests but also pose a high health hazard to humans. Various aspects of cockroach control including chemical and non-chemical methods have been researched on an ongoing basis. Repellents against cockroaches are considered as one of alternatives to eliminate cockroaches from dwellings and other harborage places. Attempts have been made to find and develop cockroach repellents derived from plant extracts. We evaluated 20 essential oils extracted from various plants such as basil, black pepper, eucalyptus, galanga, guava, kaffir lime, etc., for repellency against two cockroach species: the American cockroach (*Periplaneta americana*) and the German cockroach (*Blatella germanica*). The experiments were carried out against adult cockroaches in the laboratory. It was found that the tested essential oils provided various degrees of repellency against cockroaches, ranging from 40 to 100%. The essential oil extracted from kaffir lime leaves showed excellent activity, manifesting 100% repellency against both cockroach species. The essential oils providing high degrees of repellency (>80%) were subsequently formulated as cockroach repellents and tested against cockroaches in the field. Data from both laboratory and field tests will be presented.

Recent trends in louse control in the USA

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Since the advent of permethrin resistance among head lice (*Pediculus h. capitis*) in the 1990's in the USA, the hegemony of permethrin and synergized-pyrethrin products has given way to a more diverse array of louse control products. Many of the new products use 'natural' ingredients since consumers are anxious to avoid applying insecticides to their chilen, but efficacy is often questionable. Products for nit removal, for repelling lice and for louse detection are all available. The synergized-pyrethrins and permethrin-based products still have a dominant position. New active ingredients and novel technologies have been, and are, being evaluated for louse control – information on most are not yet publicly available. Products now on the market are discussed. FDA requirements are summarized Market size and product market-share are estimated. Future trends are predicted.

The *Simulium ochraceum* (species complex) (Diptera: Simuliidae) and its distribution in relation to onchocerciasis transmission in Latin America.

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Simulium ochraceum is the primary vector of the disease in Central America and it could be responsible for sporadic transmission of the disease in mainland Ecuador. It consists of three cytotypes which probably have different biting behaviour and susceptibility to the parasite. The morphological variation of this species along its distribution range and its rôle in onchocerciasis transmission are here discussed. The female of *S. ochraceum s.l.* is recognised by thorax orange with 1+1 white pruinose, sub-median cunae (light source anterior). Within the complex, variation in the length of white pruinose vittae and coloration of postnotum and legs occur. *Simulium ochraceum s.l.* shows marked differences in its biting behaviour, being anthropophilic in Central and South America and largely zoophilic in the Caribbean Islands. It bites man occasionally in Brazil and Venezuela. The use of integrated taxonomic techniques is suggested to discover the meaning of morphological variations in terms of cytotypes. The host capacity of *S. ochraceum s.l.* is low because of the presence of cibarial teeth, but it is an efficient vector in hyperendemic areas for the disease. Other factors affecting the vector capacity of this species in the transmission of onchocerciasis in Latin America are discussed.

Emergence of epidemic dengue in Kerala State, southern India, with an account of establishment and dominance of the vector mosquito *Aedes albopictus* (Skuse)

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Dengue (including dengue hemorrhagic fever and dengue shock syndrome) has emerged as a major health hazard in India in recent decades as it is being increasingly reported in epidemic proportions from even States hitherto largely *terra incognito* for epidemic dengue, e.g. Kerala State in south India. Several focal outbreaks of dengue fever have been reported from Kerala in recent years, originating in the sylvatic and mountainous terrains of Kottayam, Idukki and Ernakulam, and spreading to coastal districts. The enormity of problem is gauged from the fact that the incidence hiked steeply from merely 14 cases and 4 deaths in 1997 to 3377(241-fold increase) and 66 (>16-fold increase) by November, 2003. While the principal dengue vector, *Aedes aegypti*, was insignificantly prevalent in density mostly along the coastal belt urban areas, *Ae. albopictus* has dominated in prevalence (71.2%), followed by *Armigeres subalbatus* (20.7%), *Aedes aegypti* (4.4%) and *Culex uniformis* (1.5%) amongst all the 22 species under seven genera occurring in the State. Twice the dengue virus had been detected in *Ae. albopictus* adults reared from the field-collected immatures from Western Ghat region. *Aedes aegypti* is, however, yet to be incriminated from Kerala State.

The truth is out there: application of new technology for the understanding and surveillance of mosquito-borne arboviruses

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Since 1995, Japanese encephalitis virus (JEV) has emerged as an annual threat to northern Australia. To monitor for future incursions of JEV, a sentinel pig programme was established in the Torres Strait and on Cape York Peninsula. While this system has been successful in detecting the virus, it is not ideal: deployment of the pigs is expensive, dangerous to handlers, and, as pigs are the primary amplifying hosts of JEV, they could serve as a source of virus. We outline the development of a mosquito-based sentinel system for the detection of JEV activity in remote areas of northern Australia. This approach is based on application of new generation mosquito traps, such as the propane-powered Mosquito Magnet™, for collection of mosquitoes, and a real time TaqMan RT-PCR for specific detection of viral RNA. In a field trial of the system, it was demonstrated that a single JEV-infected mosquito could be detected in pools of <5000 mosquitoes, maintained for <14 days in tropical conditions. Since 2002, 10 mosquito pools have been positive for JEV at the same time sentinel pigs have seroconverted to the virus. The relative efficacy of the mosquito-based system for detection of JEV and other arboviruses will be discussed.

The New Year's party that went terribly wrong – A case study on the influence of wrapping on decomposition of a body.

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In this study, an example of unusual disposal of remains is presented with comments on the changes in the resulting patterns of arthropod invasion. A young female disappeared the night of 31st December 2001, after a New Year's party. The evening of 12th January 2002 a suspect was arrested and the body was found. The body was in an advanced stage of decomposition, almost skeletonized. The insects present were late third instar *Chrysomya albiceps* larvae, a few adult *Dermestes maculates* and a few adult Piophilidae. These insects did not correspond to the stage of decomposition. Diptera pupae were also expected. The question arose whether the body might have been kept for a few days, excluding insects, before disposal. It was later established that the body was wrapped in a cotton sheet before it was left in the field. The crime scene was reconstructed, using two pig carcasses, the one wrapped in a sheet and the other unwrapped. Decomposition and insect succession of the two carcasses were monitored and compared. The insects present on the carcasses were the same, but the wrapping of the carcass slowed down the decomposition process, although colonization of the two carcasses occurred at the same time.

The colonization of buried remains at different depths

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A body exposed in the environment represents a trophic resource for necrophagous organisms. Faunal succession and free access to the corpse are influenced by environmental factors and by the state of the body remains. Instead, in the case of buried corpses, few organisms are able to reach and process these remains. To study which insect species are responsible for underground degradation, we performed experiments by burying some boxes with meal at different soil depths. Two experimental areas in NE Italy were chosen: one in a hilly landscape without agricultural activities, the other in a zone with high human exploitation. In these sites both grassland and deciduous forest were tested. The buried boxes were collected at different times and insect specimens were identified. Diptera belonging to 8 families were found: Stratiomyidae, Phoridae, Sphaeroceridae, Piophilidae, Anthomyiidae, Muscidae, Sarcophagidae and Calliphoridae. The most abundant being the Phoridae, with *Diplonevra funebris* the most abundant species (80% of the total). Calliphoridae and Sarcophagidae were present only in the non-buried boxes. Several Coleoptera were also collected: Staphylinidae were found at all the depths, both as adults and larvae, while Silphidae and Dermestidae were collected only in the non-buried boxes.

Bionomic of ***Anopheles stephensi*** **Liston in a malarious area, Hormozgan province, southern Iran, 2002**

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A total of 25000 malaria cases has been reported from Iran. From which Hormozgan province has a higher API comprising 30% of total cases. *Anopheles stephensi* is an important malaria vector in Iran. This species has two activity peaks, one in spring and another in autumn in the region. Mosquitoes of this species prefer to rest and feed indoors. Larval habitats are quite different. All three known biological forms of this species; type, intermediate, and *Mysorensis* occur in the province. Insecticides susceptibility tests showed susceptibility to bendiocarb, propoxur, malathion, fenitrothion, deltamethrin, permethrin, cyfluthrin and lambdacyhalothrin, but resistant to DDT and low level of tolerance to dielin. Larvicidal activity of malathion, fenitrothion, temephos and chlorpyrifos at diagnostic dose exhibited susceptibility to all larvicides except to fenitrothion. The irritability tests to pyrethroid insecticides showed a high level of irritancy to permethrin and lambda-cyhalothrin, but low irritancy to cyfluthrin and deltamethrin.

Identification of Australian carrion-breeding blowflies using multiple gene regions

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"Carrion-breeding blowflies have well recognised forensic importance. Their period of development on a corpse can help indicate the time elapsed since death. The applied importance of these flies makes their reliable identification a necessity. Mitochondrial DNA markers may help identify immature blowflies whose identity is difficult or impossible to confirm morphologically. DNA recognition of Australian blowflies poses a special challenge, because their relatively recent speciation makes it necessary to focus on highly variable gene regions for their molecular separation. Thirty-four blowfly species from southern and north-eastern Australia were included in our study, representing almost all species currently known or suspected to be carrion-breeders. Phylogenetic analyses of DNA sequences from the mitochondrial genes COI, COII and ND4 were conducted using parsimony, maximum likelihood and Bayesian methods. In general, these genes provide a powerful tool for the identification of the immature stages of Australian carrion-breeding blowflies. However, in some cases mtDNA sequence divergence is very low, even though statistical support for the separation of individual specimens may be high. Since this leaves open the potential for misidentifications, we have also investigated the applicability of variable nuclear gene markers, the ITS1 and ITS2 regions, to the molecular identification of very closely related species."

Emerging Vectors in Urban Wetland Ecosystems

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The use of constructed treatment wetlands for water quality improvement, water reclamation, wetland habitat supplementation, and other goals is increasing worldwide. Man-made wetlands can provide a cost-effective alternative to conventional water treatment facilities for municipal wastewater and are one of several technologies being used to improve the water quality of urban/suburban storm water runoff. A drawback of man-made wetlands used to treat municipal and storm water in areas of rapid human development is the juxtaposition of developmental sites for pathogen-transmitting and pestiferous mosquitoes with human development and peridomestic reservoirs of the causative agents of disease. Management practices and design features thought to be important for water quality improvement often enhance mosquito production. Vector control activities can be impeded by the management practices used to fulfill simultaneously the diverse goals for multipurpose constructed wetlands. The production of native mosquito species which are known to be vectors of the causative agents of disease remains a concern at man-made wetlands and at many best management practices for treating non-point pollutants in the urban environment. The introduction of novel pathogens and vectors into urban landscapes containing treatment wetlands creates new challenges for agencies charged with protecting public health.

The response of mosquito populations to conservation, rehabilitation and construction of urban wetlands at Sydney Olympic Park, NSW, Australia.

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Homebush Bay once contained diverse, yet degraded, estuarine and freshwater wetlands that were either removed or rehabilitated in conjunction with the Sydney Olympic Park urban redevelopment. In addition, extensive freshwater wetlands were constructed for water storage, waste-water treatment, wildlife conservation and aesthetic enhancement. Over 50 mosquito species belonging to 8 genera have been identified from the area, and the potential increases in nuisance biting and public health risks following redevelopment have been a major concern for local authorities. Poor tidal flushing of the estuarine wetlands resulted in unusually large populations of the saltmarsh mosquito, *Ochlerotatus vigilax*, a known virus vector and biting pest, but populations have declined following the commencement of pesticide application and wetland remediation programs. *Culex annulirostris* and *Coquillettidia linealis* are two freshwater pest/vector species that were abundant but declined with the redevelopment. Although the constructed freshwater wetlands were designed to minimise mosquito populations, they have become more productive with increasing vegetation density. Additionally, domestic species *Ochlerotatus notoscriptus* and *Culex quinquefasciatus* are expected to increase with the further residential development. This presentation will provide commentary on the response of mosquito fauna to changes in estuarine and freshwater habitats influenced by the Sydney Olympic Park urban development.

Using vector-borne disease outbreaks to guide ecosystem interventions.

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There is close association between ecosystem health and human health, exemplified by disease outbreaks that result from ecosystem disruptions. Historical examples include the massive mortality from yellow fever and malaria during construction of the Panama Canal, and more recently the emergence of Lyme disease in ecosystems with decreased biodiversity. In order to decrease the burden from such vector-borne diseases, it is necessary to identify disrupted ecosystems for early remediation. Human health is generally subject to the most widespread and comprehensive surveillance compared to other proposed 'indicators of ecosystem health'. Thus we suggest that human disease outbreaks are one of the most useful and practical bioindicators of the health of an ecosystem. Indeed, many subtle ecosystem disruptions are often only identified as a result of detailed epidemiological investigations following an anomalous increase in human disease incidence detected by routine surveillance mechanisms. Outbreak data can act as a pivotal warning system for ecosystem disruption and can be used to identify interventions for preservation of ecological (and ultimately human) health. Using this approach, appropriate strategies for intervention and remediation can be introduced at an earlier stage than would be possible based solely on environmental monitoring, thereby improving ecosystem health and human health concurrently.

Genotyping human and other vertebrate DNA from maggot gut contents: practical considerations

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Genetic analysis of maggot gut contents may aid a death investigator. We performed experiments aimed at defining procedures and conditions that will allow such an analysis to succeed. Calliphorid larvae were fed on beef, pork, rat, dog, cat, sheep, rabbit, deer, and human tissues. Larvae were then harvested at a range of developmental stages. Conventional forensic DNA typing procedures were performed on larval gut DNA extracts. The effects of maggot development, specimen preservation technique, larval starvation, and transfer of a feeding larva to a new food type, on recovery of the correct food genotype were examined. PCR amplification involved new cytochrome b primers specifically for vertebrate and not insect DNA, and a commercial STR kit. 20% bleach destroyed surface contamination without interfering with the analysis of internal tissues. Larvae manually removed from food or naturally postfeeding were unsuitable for this analysis after 24 hr off the food. For larvae transferred between food types, both the original and second food genotype could be recovered 46 hr following transfer if specific primers were used. Our methods failed with formalin-preserved specimens. However for all other preservation strategies STR and mtDNA types were obtained from at least some specimens after six months of storage.

Cellular and molecular immunology of the tick-host interface: implications for pathogen transmission

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Complex immunologic interactions occur at the tick-host interface. Tick feeding stimulates host innate and specific acquired immune responses to limit blood meal acquisition. In response to host defenses, tick saliva contains molecules that counteract host immunity. Tick infestation suppresses lymphocyte proliferation, polarizes/modulates cytokine responses, reduces adhesion molecule/integrin expression, modifies antigen presentation, and inhibits cytokine action. The cellular and molecular bases for these phenomena are being defined and multiple diverse pathways for achieving the same end point of immunomodulation are being characterized. The earliest stages of specific T-lymphocyte responses are being analyzed. Genomic and proteomic approaches are used to identify the molecules responsible for modulation of host immune defenses. Tick salivary gland gene expression is being characterized during the first few hours of feeding, and salivary gland molecules of interest are being cloned, expressed and their biological activities identified. Emerging from these studies is a picture of a web of interactions essential for successful blood feeding and transmission of infectious agents that is far more complex than initially envisioned.

Genomics and the development of vaccines against ticks and tick borne disease

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Whole genome sequences of major tick borne disease organisms are available or in progress. Sequencing of the first tick genome will commence soon. Host microarrays allow the dissection of the response to tick or pathogen infestation in a novel way. However, analysis of this deluge of information remains problematic. The rate-limiting step in vaccine development typically remains the assessment of candidate antigens and delivery systems in animals. Genomic information to be useful must be filtered to select a small number of candidates from a plethora of choices. Experience with *Boophilus microplus,* other tick species as well as *Anaplasma marginale* suggests that few highly effective antigens have yet been discovered and the reason for the efficacy of those remains obscure. Other, often less efficacious antigens identified to date fall principally into a few biochemical classes while many other potential antigen classes remain unexplored. The effective use of genomic information will demand a greatly enhanced understanding of the biology of vaccination than is currently available.

cDNA cloning, characterization and preliminary vaccine effect analysis of two *Rhipicephalus haemaphysaloides haemaphysaloides* cathepsin L-like cysteine proteinases

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Two cathepsin L-like cysteine proteinase genes were cloned from the tick, *Rhipicephalus haemaphysaloides haemaphysaloides*, named as RhcA and RhcB. The full length of RhcA is 1168bp, encoding a 332 amino acid residue polypeptide; the full length of RhcB is 1153bp, encoding a 335 amino acid residue polypeptide. The consensus amino acid motifs presence in both deduced amino acid sequences and both genes show high sequence homology to other cathepsin L-like cysteine proteinase genes, so they were identified as members of the cysteine proteinase gene family. Expression analysis by RT-PCR revealed that RhcA and RhcB were expressed differently in different stages of tick development. Western blot proved the anti-saliva serum can recognize the *E.coli* expressed recombinant proteins. The immunization test of recombinant against tick infestation showed the two molecules exhibited partially vaccine effects in attachment rate and engorgement rate of adults. GenBank accession No. AY336797 and AY336798 are for RhcA and RhcB respectively

Seasonal studies on the influence of clothing and wrapping on carcass decomposition and arthropod succession.

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Six pig (*Sus scrofa* L.) carcasses, three wrapped and clothed carcass and three wrapped and naked, were sampled i) daily ii) after five days and iii) after ten days. Two more carcasses, one naked and not wrapped, and one clothed but not wrapped were sampled daily as controls. Differences in decomposition between the clothed or wrapped carcasses included the distribution and size of maggot masses and significant delay in ying out of the carcasses for all seasons. The maggot masses were primarily dominated by *Chrysomya marginalis* (Wiedemann) and *C. albiceps* (Weidemann), in autumn and summer. *Chrysomya chloropyga* (Weidemann) and *Calliphora vicina* Robineau-Devoidy in winter, with *C. chloropyga* and *C. albiceps* in spring. There was no delay in oviposition except in winter where a five day delay occurred on the wrapped carcasses. Extremely high internal temperatures in the warmer seasons caused significant maggot mortality on the wrapped carcasses. The predation of *C. marginalis* maggots by *C. albiceps* maggots was observed in autumn. In summer, maggot masses of these species were observed in separate groups. The coleopteran community was dominated by *Thanathopilus micans* L. larvae, succeeded by *Dermestes maculatus* De Geer, larvae. Adult *D. maculatus* and *Necrobia rufipes* De Geer, were present on the carcasses throughout all seasons.

Ectoparasites and arthropod-borne parasites of brown rat, *Rattus norvegicus*, **in Ahvaz, southwest Iran**

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Arthropods play an important role in the transmission of infectious agents to human and animals, such as rodents. Ahvaz, southwest Iran, is encountered with high density of rodents, especially *R. norvegicus*. In this study, 72 *R.norvegicus* were captured from different sites of Ahvaz, using live traps. Their ectoparasites were collected and identified. Thin and thick blood smears were prepared. After dissection of rats, their different organs were examined for any endoparasites, applying specific techniques. In general ectoparasites comprised 2 species of flea, *Xenopsylla astia* and *X. nubica*, 1 species of louse, *Polyplax spinula* and 1 species of mite *Laelaps nuttalli*. Among endoparasites, 9 species utilize arthropods as vector or intermediate host in their indirect life cycles. These included 1 species of protozoan parasite, *Trypanosoma lewisi* and 8 species of helminth parasites. The potential risk of each of these identified species to the health of human and animals will be discussed.

Applications of GIS and remote sensing for surveillance and control of mosquito borne diseases

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The risk of transmission of mosquito borne diseases depends on the spatial distribution of vector species in relation to the distribution of human hosts and, in case of mosquito-borne zoonoses, the distribution of vertebrate reservoir hosts. These distributions and temporal changes in these spatial patterns depend on environmental and climate/weather conditions. Satellite imagery provides means to gather vast amount of landscape/landcover and meteorological data that are proximate determinants of vector distribution and risk of disease transmission. Geographical information systems allow for the integration of environmental, entomological, epidemiological and demographic data and provide a framework for surveillance and control on scales ranging from local health departments and mosquito control districts to national and international programs. Products of applications of these tools and quantitative analysis of spatial data include data management tools, disease risk maps, and decision making tools for location and time of interventions. Examples from studies in the U.S. Israel, Trinidad and Kenya will be presented, including malaria cases in relation to *Anopheles* larval sites in Trinidad and Israel, dengue and dengue haemorrhagic fever in Trinidad, eastern equine encephalitis outbreaks in Michigan, USA, and La Crosse encephalitis transmission and the effect of introduction of *Aedes albopictus* in Illinois, USA.

Suppression of dengue transmission by application of integrated vector control measures at sero-positive GIS-based foci: demonstration of a cost-effective model

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A serological survey of approximately 2,000 primary school chilen from six schools in Chachoengsao, Thailand, was performed at the end of the peak dengue transmission period. GIS analysis of sero-positive cases was carried out to determine transmission foci in high IgG-IgM positive villages. Vector control implementation was conducted in the foci and also within 100 meters around the foci, including schools attended by chilen in the treated areas, with collaboration from local public health and school authorities. Vector control measures included source reduction by routine garbage pickup organized by local government, the use of locally made screen covers, a combination of local Bti and *Mesocyclops*, and locally made lethal ovitraps. Implementation of vector control measures in the foci was continued until the end of the rainy season. Vector control efficiency was monitored using both entomological and serological parameters. Results showed a significant reduction of dengue vectors in treated areas when compared to untreated areas. Despite overall higher dengue incidences in the second year when compared to the first year, IgG-IgM positive students in the treated areas decreased from 13.46% to 0% while those from untreated areas increased from 9.43% to 19.15%.

Novel mosquito traps: can they control moquitoes

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A diversity of commercial traps for mosquito control have become available during the past five years. Most are marketed to homeowners. Many models have been evaluated in a large outdoor screened enclosure against two species (*Aedes aegypti* and *Ochlerotatus taeniorhynchus*) of laboratory reared mosquitoes to determine their relative efficacy. Several models were selected for field testing against natural populations. Ongoing research is trying to answer the question: Can mosquitoes be trapped out? Different management strategies are being evaluated, e.g. control potential on isolated islands versus urban/suburban residential settings. In urban/residential settings traps are being evaluated as a protective barrier around a city block and in individual yards. Preliminary results indicate that protective trap barriers provide excellent control (>90%) on isolated islands, but only moderate (50% reduction) control in urban/suburban areas. A single trap in a single yard provided no measurable mosquito control.

The mode of action of deet, SS220, and Bayrepel® arthropod repellents against *Aedes aegypti*, *Anopheles stephensi*, **and** *Phlebotomus papatasi.*

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Laboratory bioassays were conducted with human subjects using six-celled K&D modules and the known arthropod repellents, N,N-diethyl-3-methylbenzamide (Deet), (1S, 2'S)-methylpiperidinyl-3-cyclohexene-1-carboxamide (SS220) and 2-(2-hydroxyethyl)-1-piperidine carboxylic acid 1-methylpropyl ester (Bayrepel) against *Aedes aegypti* (Linnaeus), *Anopheles stephensi* Liston mosquitoes and the sand fly, *Phlebotomus papatasi* which are vectors of human diseases. When five females of each species were simultaneously exposed to one half repellent-treated (4.8×10^{-2}µmol/cm^2) and one half untreated skin beneath the opening of a 3x4 cm cell, they bit exclusively on untreated skin. When treated and untreated skin was covered with cloth to preclude tactile contact with skin surfaces, the insects bit only through cloth covering untreated skin. The tests proved that each species was exquisitely capable of avoiding the compounds in their close-range feeding behavior and this was accomplished by olfactory chemical sensing. When cloth, untreated and repellent treated, was placed over the skin all species avoided the compounds and fed through the cloth only where repellent was absent. In the presence of any the three repellents within a cell, biting responses were reduced by about 50% relative to controls. Thus, besides inhibiting feeding on repellent-treated skin, the compounds also exhibited spatial-repellent activity.

Key issues in advancing the sterile insect technique for African malaria vectors

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Pilot projects that assessed the feasibility of the sterile insect technique (SIT) against Anopheles mosquitoes were conducted in El Salvador in the 1970s. Though on a relatively small scale, these trials demonstrated that sterility could be induced in a wild mosquito population through releasing (chemo)sterilized males. Since then, major scientific advances merit a renewed look at SIT and its applicability in the African context. We describe the R&D phase currently underway in Sudan and Austria, to develop appropriate mass-rearing systems and sterilization procedures. The salient features of such releases, also in relation to the use of transgenic approaches, will be discussed. These include colonization and mass rearing, (genetic) sexing, mosquito fitness and release mechanisms.

The influence of burning on the decomposition and insect succession of pig carcasses.

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Recent casework raised the question whether burning has an influence on the decomposition and insect succession of bodies. This was investigated by dousing three pig carcasses with different volumes of petrol and burning them to different degrees (Slight = 1.5L Medium =5L, Heavily = 10L). The insect succession onto these carcasses was compared to a control carcass. The control carcass decomposed at the fastest rate with the heavily burnt carcass at the slowest rate. Adult insects comprised the following: At the control carcass Diptera (60.8%), Coleoptera (29.9%) and Hymenoptera (8.8%). At the slightly burnt carcass Coleoptera (55.8%), Diptera (38.6%) and Hymenoptera (5.4%). The dominant orders at the medium burnt carcass were Coleoptera (38.5%), Diptera (33.3%) and Hymenoptera (28%). At the heavily burnt carcass Coleoptera (51.4%) and Diptera (28.6%) were dominant. Hymenoptera (18.7%) did not have a direct influence on decomposition, since they functioned as predators. *Chrysomya albiceps* was the dominant Calliphoridae species at the control and medium burnt carcasses and *Chrysomya marginalis* at the slightly and heavily burnt carcass. Representative samples of larvae collected during decomposition revealed that *C. albiceps* larvae preferred to feed underneath the carcasses, with *C. marginalis* larvae feeding mostly inside and at the top of the carcasses.

The study of excito-repellency phenomenon of *Anopheles stephensi* exposed to nets impregnated with cyfluthrin

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The excito-repellency phenomenon is occurred in the vector population following the application of definite insecticides. Based on the field observation, in relation to residual spraying operation, the excito-repellency phenomenon may led to behaviouristic changes of the mosquitoes as diversion either to new hosts or other unsprayed shelters. This occurrence may lead to failure in control program. At these conditions, the mosquitoes leave the sprayed houses to either unsprayed or outdoor shelters before picking up the lethal dose. The result of this investigation will be useful for planning of the control program in the country. For the first time, the standard design of excito-repellency (ER) test box was made of aluminum. The laboratory test was carried out with cyfluthrin treated nets against *An.stephensi* (Beech strain). The result showed that the tendency of the mosquitoes to long resting on the netting of exposure chamber was decreased at high dosage of insecticide, and tried to leave the exposure chamber toward the exit trap. The mean of the feeding rate was least at higher dosage in comparison of lower one.

Human behaviors and environmental factors elevating the risk of exposure to *Ixodes pacificus* nymphs in a hardwood forest

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The nymph of the western black-legged tick, *Ixodes pacificus*, has been implicated as the primary vector of the Lyme disease spirochete, *Borrelia burgdorferi*, in northwestern California. In 2002 and 2003, six human behaviors and environmental factors were evaluated as potential risk factors for acquiring nymphal ticks in a dense woodland in this region. The prevalence of *B. burgdorferi* and the causative agents of human anaplasmosis (*Anaplasma phagocytophilum*) and human monocytic ehrlichiosis (*Ehrlichia chaffeensis*) also were determined in nymphs that had been collected from subjects or by agging leaf litter. Time-adjusted tick-acquisition rates demonstrated that sitting on logs was the riskiest behavior followed, in descending rank, by gathering wood, sitting against trees, walking, stirring and sitting on leaf litter, and sitting on litter. The number of ticks acquired appeared to be unrelated to the type of footwear worn. Nymphal ticks were active throughout the daytime over a wide range of micrometeorologic conditions. Overall, about 3-4% of nymphs were infected with *A. phagocytophilum* and *B. burgdorferi*, regardless of the collection method, versus none with *E. chaffeensis*. Co-infection with *B. burgdorferi* and *A. phagocytophilum* was detected for the first time in this life stage of *I. pacificus*. The public health implications of these findings will be discussed briefly.

Genetically modified mosquitoes: a real world view

GC Lanzaro ADDRESS

The concept of controlling mosquito-borne diseases via the development and implementation of genetically modified strains has been under serious consideration since the early 1990's. Although this idea has been met with great enthusiasim in some quarters there remains substantial skepticism in others. The general concept and progress toward the development of such a strategy will be presented. Numerous roadblocks to the development of a GM mosquito plan remain, including (i) lack of gene drivers, (ii) lack of robust effector molecules and mechanisms to manipulate their expression, (iii) poor efficiency of transgenesis, (iv), and (v) no consideration of logistical problems associated with implementation in the field. These will be presented and prospects for overcoming them discussed. Aspects of the ecology and population genetics of targeted species will be discussed as they relate to the successful release and establishment of GM mosquitoes in nature.

Reverse chemical ecology: chemical prospecting of mosquito attractants and repellents

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For a few decades, chemical ecologists have been utilizing insect antennae as the sensing element or biodetector, in electroantennogram (EAG) and gas chromatography-electroantennographic detection (GC-EAD) assays for the identification of minute amounts of pheromones and other semiochemicals. The "molecular" equivalent of these electrophysiology-based "short cut bioassays" is the screening of attractants and repellents based on their binding affinities to odorant-binding proteins (OBPs). OBPs assist hydrophobic semiochemicals to cross an aqueous barrier in the insect sensilla (sensillar lymph) and reach odorant receptors (ORs). Both OBPs and ORs contribute to the high specificity of the insect olfactory system by forming two layers of filters. Although binding per se does not necessarily imply a physiological function, test compounds that do not bind to OBPs can be eliminated from further behavioral studies as they will never "see" the receptors. Reverse chemical ecology is a protein-based approach that requires the full identification of OBPs, cDNA cloning, functional expression of OBPs, and binding assays. In this presentation, we will discuss the latest development of a NIH-funded project* aimed at the development of better attractants and repellents for *Culex* species, the principal vectors of West Nile Virus.* NIH-National Institute of Allergy and Infectious Diseases (1U01AI058267-01).

Developing methodologies for a fungal biopesticide against *Boophilus* microplus, the cattle tick

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The Queensland department of primary industries and fisheries has been investigating fungal biopesticides for livestock ectoparasites as part of a wider strategy to reduce the reliance upon chemicals in livestock industries. Fungal biopesticides have been available for the control of agricultural and horticultural pests for over thirty years and methodologies and accepted protocols for the development of these fungal biopesticides are now well established. Current work with the cattle tick has shown that livestock pests present additional challenges, some of which are due to the inability to conduct *in vitro* assays on obligate parasites, the cost of *in vivo* experiments using animals and the different environments the fungus must operate in when invading the parasitic and free-living phases. In addition, the development of formulation and application methods suitable for penetrating an animal's hair coat and delivering a lethal dose of spores to ticks present further challenges. Solutions to these problems have sometimes been obvious and straightforward but mostly have been arrived at slowly and painstakingly. We have concluded that the development of methodologies for fungal biopesticides of livestock ectoparasites is a multidisciplinary exercise requiring a thoughtful balance of appropriate *in vitro* bioassays with on-animal investigations.

Increased densities of vector mosquitoes in anthropogenic environments in the Kapiti region in New Zealand and potential ecological mechanisms as to why

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Landuse changes, including deforestation, agriculture and urbanisation, have coincided with increases in mosquito-borne diseases worldwide. Anthropogenic environmental changes can alter mosquito populations by modifying the characteristics of breeding habitats, but we still poorly understand the ecological mechanisms involved. Mosquito breeding habitats were sampled in native forest, pastureland and urbanland in the Kapiti region, New Zealand, and 17 environmental characteristics were measured of each. Higher immature mosquito densities were collected from mainly artificial containers in urbanland and pastureland compared to mainly ground pools in native forest. Nine environmental variables were significantly different between landuses but none of these were significantly related to mosquito density. Principal components analysis partitioned environmental data into seven ecological types. One ecological type was significantly related to both landuse and mosquito density, and positively associated with ammonia, bacteria and dissolved organic carbon (DOC), and negatively associated with nitrate and predatory damselfly larvae. The findings of this study support those from prior surveys in warmer climes of greater vector disease risk in anthropogenically-modified environments because of compromised ecosystem integrity. Unlike most previous field-based work, this study also suggests important mechanisms involved; pinpointing mainly bacteria, DOC, nitrate and damselfly larvae as acting in combination to regulate vector mosquito density.

The final word on the genetic, evolutionary and taxonomic relationship of *Pediculus humanus* and *P. capitis*.

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Body lice are thought to have diverged from head lice when humans began to wear clothes. This recent evolutionary divergence and incomplete lineage sorting has complicated assessments of whether head lice and body lice are separate species. We used microsatellite markers to test lice from hosts that were infested with head and body lice simultaneously, to see if the head and body lice had interbed. Our results showed that head and body lice had not interbred on a host. Assignment tests revealed that lice that had migrated among hosts always moved from head to head, and body to body, but never between heads and bodies. We conclude that head and body lice are separate species. We speculate that head lice are descended from the lice of neanderthals and modern humans, and that body lice are descended from the lice of modern humans that left Africa about 100,000 ya.

The search for natural repellents from human beings against mosquitoes

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Blood sucking insects are differentially attracted to human beings. This is likely to be caused by differences in the semiochemical profiles of individual human hosts. Despite substantial research into insect-human interactions, the responsible semiochemicals have not yet been identified. An explanation for this difference is that unattractive individuals either lack attractant chemical compounds or may produce compounds that 'mask' the activity of attractants. A special air entrainment technique was developed and used to collect volatile chemicals from human volunteers, and the extracts analysed by gas chromatography (GC), coupled GC-electroantennography (GC-EAG) and GC-mass spectrometry (GC-MS). For the first time electrophysiological responses of female yellow fever mosquitoes, *Aedes aegypti* to volatiles from air entrainment extracts were recorded, and a number of electrophysiologically active compounds were identified. Previous behavioural studies have confirmed the differential attractiveness of volunteers to biting insects. A quantitative and qualitative analysis of compounds within the extracts, from individuals known to be differentially attractive to biting insects, has revealed significant differences in chemical profiles. The identification of behaviourally active compounds could lead to new and improved control technologies, whereby 'masking' compounds could potentially be incorporated into new, safe and natural repellents against biting insect pests.

Anopheles arabiensis **at the edge of its distribution in Northern State, Sudan**

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Northern state, Sudan is mainly desert, rainfall is rare and most life is found close to the Nile. *Anopheles arabiensis* is the only anopheline and malaria vector found in the area in recent decades. It is present along the Nile downriver to about 200 km south of the Egyptian border, where poor terrain and control efforts prevent its spread further north. Initial results are presented from field and molecular studies to determine the population structure and dynamics of the species and related environmental factors. The receding Nile following the flood contributes most to sustaining *A. arabiensis*. Larval breeding sites can be found on low lying riverbank, ied-up inlets, or channels around islands, through most of the year. Land use is mainly associated with irrigation, but this does not appear to contribute greatly to anopheline numbers, however the extent of human presence and activities do increase the potential for anopheline breeding. Upriver from the fourth cataract and downriver from the third cataract, the terrain is rocky desert and less suitable for both humans and mosquitoes. The *An. arabiensis* population is thus semi-isolated within an approximately 700 km corridor a few kilometres wide and breaks down further into pockets depending on particular river features.

Control of biting midge (Ceratopogonidae) attack on residents of a marine canal estate in Redcliffe (Australia) by residual barrier treatment of Bistar 80SC® to ornamental garden plants in house yards

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Biting midge (Ceratopogonidae) breeding in marine sediments associated with engineered canals in a residential community in Redcliffe Australia have been responsible for considerable biting attack and nuisance. Limitations on approved applications of larvicides in breeding sites has historically restricted control to personal protection, midge screening and education of residents. In October 2003 – Feb 04, Redcliffe City Council funded a trial to test the efficacy of Bistar 80SC® (FMC Chemicals Pty Ltd) as a 0.1% AI (Bifenthrin) residual application to ornamental garden vegetation and dwelling external walls. Eight paired randomised treated and untreated premises were assessed for differences in biting midge exposure. The assessment was undertaken using CO_2 and Octenol baited light traps placed in treated and untreated yards. A self administered questionnaire scoring individual household daily experience of biting midge activity was also used. Untreated control premises received a placebo application of water alone to control bias in the resident questionnaire. Analysis of difference in light trap collections between treated and untreated showed a 76.4% reduction in midge (primarily *Culicoides molestus*) from treated premises. Resident survey results indicated occupants of treated premises experienced generally favourable biting midge conditions while residents from untreated homes experienced fluctuating exposure and discomfort corresponding to lunar emergence cycles.

Aerial efficacy assessment of Anvil 10+10 against *Anopheles quaimaculatus*

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Aerial ULV applications of Anvil 10+10 were conducted against caged, field-collected *Anopheles quaimaculatus* adults using Beecomist® 360A rotary atomizers. Standard and optimized applications were conducted at 0.62 fl oz (0.0036 lbs a.i.) per acre using stacked passes 3,500 ft and 5,000 ft upwind of the target site. Efficacy of Anvil 10+10 and impact of wind velocity at flight altitude (150 ft) were assessed by bioassay and analysis of collected droplets.

The potential role of aquatic insects in the transmission of Buruli ulcer disease in Africa

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Buruli ulcer is the third most frequent mycobacterial disease in humans, next to tuberculosis and leprosy, often causing serious skin ulcerations, bone deformities, and disability. Unlike TB and leprosy, Buruli ulcer has emerged amatically since the 1980's, with increasing numbers of cases being reported from at least 31 countries, mostly in tropical and subtropical areas, including Africa. Although poorly understood, the epidemiology of Buruli ulcer associates the resurgence with people who live and/or work close to wetlands, especially slow flowing (riverine) or stagnant water bodies (marshes, swamps), often created as a result of some human environmental disturbance. The disease is not known to be contagious between humans, and epidemiological studies have not yet established natural reservoirs or modes of transmission. However, it was recently demonstrated experimentally that *M. ulcerans* could survive and multiply exclusively within salivary glands of aquatic bugs (Naucoridae: *Naucoris* sp.) and they were able to transmit *M. ulcerans* to mice by their bites, suggesting a possible mechanism for human infection. The organism has been found in samples of water, biofilm and detritus. A possible trophic scenario for the natural mode of transmission of *M. ulcerans* is presented based on preliminary work in Ghana by the authors.

Efficacy of a Spinosad suspension concentrate formulation for controlling chewing and sucking lice on cattle under field conditions

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Field studies were conducted with cattle in Wisconsin and Illinois between January and April, 2000, to determine the efficacy of 0.04% spray and 2 mg/kg pour-on treatments of a spinosad suspension concentrate formulation against chewing and sucking lice. Positive control substances included in the study were Co-Ral® EC (5.8% coumaphos) and CyLence™ Pour-on (1% cyfluthrin). Animals in the negative control group remained untreated. All lice species were visually sampled on each animal at weekly intervals after treatment, for eight weeks. Individual louse counts were summed over each examination site for each species. A repeated measures analysis using a mixed model (SAS PROC MIXED), was conducted for data pooled across both study sites. Both spinosad treatments and CyLence™ Pour-on provided excellent efficacy (98-100%) against *Bovicola bovis* over the entire study period; Co-Ral® EC efficacy opped below 90% from Weeks 6-8. The efficacy of the spinosad spray treatment against *Linognathus vituli* was >93% for the entire study and >88% for the spinosad pour-on treatment. Co-Ral® EC was >90% efficacious against *L. vituli* during Weeks 1-7, while CyLence™ Pour-on only exceeded 90% for Weeks 3-7. *Solenopotes capillatus* were found only at the Wisconsin site; all treatments were highly efficacious throughout the study period.

The effect of human age on Dengue seroconversion risk in Iquitos, Peru

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Basic models of infectious disease often assume that force of infection is independent of host age. This assumption has been shown to be violated for many directly transmitted diseases, but has rarely been examined in vector-borne systems, such as dengue. Data from DNA fingerprinting studies of mosquito bloodmeals indicate that the biting rate of *Aedes aegypti* increases with human age. If risk for dengue infection increases with human age, models assuming an age-independent risk will underestimate the proportion of the population that needs to be vaccinated in order to eliminate disease transmission. A cohort of approximately 3,000 schoolchilen and their families in Iquitos, Peru were followed for up to 5 years (1999-2003) with blood samples collected at 6-month intervals. Sera were tested for dengue virus antibodies by the plaque reduction neutralization test. The effect of human age on risk for dengue seroconversion was evaluated using a multiple logistic regression model, which also included area of residence within the city. Preliminary analysis suggests that risk increased with age and was highest in the southern region of the city. Estimates of age-specific forces of infection can be used to design an optimal immunization program.

Oviposition site selection by mosquitoes in response to drought

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Mosquitoes are vectors of many important medical and veterinary diseases. Although outbreaks of mosquito borne disease can be correlated to changes in precipitation in the very short term, precipitation rarely corresponds to mosquito abundance in the long term. Here, I present the results of a field based mesocosm experiment designed to test how water level and drought influence the oviposition behaviour of two species of mosquitoes. I used two water levels (high and low) and two drought treatments (normal and drought). Water for the drought treatments was obtained from ponds that had experienced drying for a period of a month prior to experimentation. The two mosquito species differed markedly in their oviposition response. The egg laying behavior of *Ochlerlatatus notoscriptus* was significantly greater in the drought treatments regardless of water level. However, the egg laying behavior *Anopheles annulipes* showed the opposite response with significantly more larvae present in the high water treatments regardless of whether the treatment water was normal or in drought. My results provide supportive evidence that mosquitoes may show population outbreaks following drought years and that this framework may provide a reliable way of predicting year-to–year variation in mosquito abundances based on individual species oviposition response.

Aedes aegypti surveillance in Iquitos, Peru: a 4- year test of the pupal/demographic survey

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We present results from 4 years of pupal/demographic surveys of a circuit of ≈ 6,000 houses surveyed >10 times in the Amazonian city of Iquitos, Peru. We compare traditional larval indices with pupal and adult mosquito indices. We evaluated *Aedes aegypti* production from different container classes, including those based on water management practices overtime and space. A total of 289,941 water-holding containers were characterized, of which 7.3% were positive for *Ae. aegypti*. Although, temporal and geographic variations were detected for all variables examined, consistent production patterns were observed for the lid status (lids: 32% wet containers, 2% pupal production), container location (outdoor: 43% wet containers; 85% pupal production) and method by which the container was filled with water (rain filled: 15% wet containers; 88.3% pupal production). Based on these patterns and additional targeting of containers with highly variable pupal counts we propose a targeted control strategy for Iquitos that would reduce *Ae. aegypti* populations below published but untested threshold values for Iquitos.

Screening medicinal plant extracts for effects on *Aedes aegypti* (Diptera: Culicidae) & toxicity to a non target organism

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A preliminary study was conducted to find out the effects of the extracts of 112 medicinal plant species, collected from the southern part of Thailand, on *Aedes aegypti*. Studies on larvicidal properties of plant extracts against the third and fourth instar larvae of *Ae. aegypti* showed fourteen species with high toxicity. Six plant species were comparatively more effective against the larvae at very low concentration. These extracts demonstrated no or very low toxicity to guppy fish (*Poecilia reticulata*). Three plants with promising larvicidal activity, *Mammea siamensis*, *Anethum graveolens* and *Annona muricata,* were used to study of the impact of the extracts on the life cycle of *Ae. aegypti*. The results showed that these plants affected the reproductive potential of surviving adults by reducing the number of eggs laid and egg hatchability. The first instar larvae were very susceptible to *A. muricata* and the second instar larvae were susceptible to *A. graveolens* while the third and fourth instar larvae were susceptible to *M. siamensis*. These extracts delayed larval development and inhibited adult emergence.

The mosquito burden in Kenya

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Kenya is a tropical country with a climate favourable for diverse insect life – both the harmful and the useful alike. Embu District experienced a malaria outbreak between January and February 2001 where more than 300 people were admitted at the main health facility in the District, 70 lives were lost. Malaria is endemic in Embu district with a morbidity of 60% and mortality of 25%. Expectant mothers and chilen are affected most with slide positivity of 30% and 45% respectively. Annually, each household spends around US$ 65 on malaria control and funeral expenses. Larval densities range from 30-40% with peak densities in March, April and May. Despite this, 25% of the community cannot positively identify mosquito as the malaria vector. Only 12% can correctly name the five main symptoms of malaria (fever, chill, sweating, anorexia, joint pains). The ministry of health has issued guidelines on uniform treatment and community sensitisation. Consistent use of bednets, chemoprophylaxis and indoor residual spraying are being encouraged. Plans are underway to start biological adult and larval control. This paper discusses the mosquito burden from the perspectives of larval density, host prevalence index, mortalities and morbidities, bed occupancy and the economics of vector control

Tick vector-pathogen interrelationships

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The interrelationships between tick-borne pathogens, their tick vectors and diverse vertebrate hosts, are represented by a triangle of parasitic interactions. The interactions are between (i) tick-pathogen, (ii) pathogen-host, and (iii) tick-host. In (i) the pathogen interacts with its vector, infecting and replicating within tick cells or extracellular spaces. The ability of a particular tick species to act as a vector depends on whether the pathogen can survive and overcome several 'barriers' within the tick. However, this is a simplistic view of the tick vector-pathogen interrelationship as both (ii) and (iii) influence the success of (i). This triangle of interactions is focused at the skin site of tick feeding - the initial site at which pathogens gain access to either their host or vector. For pathogens, the skin-feeding site is an ecologically privileged niche, profoundly modified by the pharmacological activities of bioactive tick saliva molecules. Exploitation of this niche is known as saliva-activated transmission (SAT). Tick saliva molecules that promote SAT have not been identified but appear to differ for different tick vector-pathogen interrelationships. Identifying the SAT factors and their mode of action offers new insights for controlling ticks and tick-borne diseases.

Argentina: veterinary forensic entomology as a potential tool against rustling

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Cattle rustling entails economic loss to cattle owners and danger to their safety, marketing of uninspected meat and hides and loss to the State from unpaid taxes. In 2003, the Faculty of Veterinary Medicine (Tandil, Buenos Aires province, Argentina) incorporated forensic entomology to a course of legal veterinary medicine aimed at fighting rustling. Preliminary field experiments showed a large proportion of hemisynanthropic insects, most of them Neotropical, which may represent the autochthonous necrophagous fauna, now displaced by synanthropic introduced species in urban environments. Colonization and development times proved longer in the arid climate of Tandil than at Buenos Aires city, where most published data were gathered. Field experiments with bovine bodies and parts are required. Hitherto, bovines have been avoided as a model because of the added complication of the ruminant digestive tract. Since most countries with rustling problems also have a pattern of introduced species displacing the original necrophagous fauna in city environments, they might benefit from knowledge obtained through these studies.

Arboviruses, mosquitoes & rNA interference

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The identification of critical arbovirus-vector interactions reveals new insights into vector competence. For instance, flavivirus and alphavirus replication in mosquitoes triggers a natural RNA interference (RNAi) response; however, viral counterdefenses probably allow RNAi evasion and establishment of persistent infections. We can enhance RNAi to dengue virus type 2 (DENV2) in *Aedes* species by administration of double-stranded (ds) RNA derived from the DENV2 genome, either by direct transfection of dsRNA, transient expression from a Sindbis virus (SINV) transducing system, or stable transformation with a plasmid that expresses a hairpin-forming transcript. RNAi is sequence-specific, is maintained by low concentrations of effector RNA, blocks accumulation of intracellular viral RNA, and results in the appearance of 21-25 nt siRNA homologous to the mRNA target. We have identified genes from the Dicer and Argonaute families in 2 mosquito genera and have used RNAi to knock down their expression to determine which genes have a role in virus resistance. We have evidence that DENV2 and some strains of SINV establish persistent infections in mosquitoes by suppression of RNAi, and are investigating mechanisms. We are identifying viral-encoded suppressors by site-specific mutations in infectious cDNA clones. Our work suggests that persistent infections in mosquitoes result from a balance between host RNA silencing and viral counterdefenses and provides new strategies for tipping the balance in favor of the vector host.

Endosymbionts & sex ratios in the human head louse

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At the turn of the 19th century the first observations of a female-biased sex ratio in broods and populations of head lice had been reported. A study by Buxton in 1938 on the sex ratio of lice on prisoners in Ceylon is still today the subject of reanalyses. However, in the last sixty years no data have been collected, especially on more normal levels of infestations. Here we report a female bias of head lice in a survey of 480 school children in Argentina. This bias is independent of lice intensity. These lice as well as lice from pupils in Britain were carrying several, related strains of the endosymbiotic bacterium *Wolbachia*, one of the increasing number of intracellular sex ratio distorters. No lice without *Wolbachia* were detected. Similar *Wolbachia* strains are also present in the pig louse suggesting that this endosymbiont might have marked influence on the biology of the whole order. The sex ratio distortion in head lice that has been reported world wide is stable over time; that is a remarkable deviation from the stability of frequency-dependent selection of Fisher's sex ratio.

Blood feeding in *Cnephia* (Diptera: Simuliidae): molecular phylogeny

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Five species of *Cnephia* were studied, three non-biting, one a mammal feeder, and one a bird feeder. Of the non-feeding species, *C. eremite* is Holarctic in distribution and specimens were examined from both Canada and Sweden. *Cnephia pallipes* is a Palearctic non-feeder and *C. dacotensis* a temperate Nearctic non-feeder. *Cnephia pecuarum* is an economically important large mammal feeder in south-central U.S. and *C. ornithophilia* a widely distributed bird feeder in southern and eastern North America. Sequence was obtained from the complete mitochondrial CO-II gene (687 bp) and partial EF1-alpha nuclear gene (1030 bp). Molecular data is discussed in relation to conventional wisdom about relationships among species based on cytological and biological information, geographic distribution and genetic diversity within species. Molecular data strongly support *Cnephia* as a monophyletic clade in the tribe Simuliini. Speciation hypotheses were not fully concordant between molecular and conventional data.

Onchocerciasis in Central America: ecology & control

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Detection of O. volvulus larvae in vector populations of black flies is of prime importance in the assessment of the effectiveness of ivermectin control programs. Detection of larvae is attained by dissection of individual flies or by PCR (amplification of an Onchocerca 150 bp repeated sequence, followed by hybridization of the PCR products with a species-specific DNA probe) from pools of flies. The PCR was compared with dissection method to estimate the infection rate (p) and the mean larval load (any larval stage) per parous fly (m) in a local vector population. Both methods produced statistically similar estimates, thus PCR is an alternative and efficient method for entomological surveillance. Sample sizes required to estimate m from p for fixed levels of precision were calculated using Mexican and Guatemalan data, assuming that k (an aggregation parameter from the larval distribution among parous flies) is known. Optimal sampling and PCR was used to evaluate the impact of ivermectin on m, p and transmission potentials in Mexico. The data suggest that substantial progress towards the goal of suppressing transmission has been accomplished. In southern Chiapas, entomological estimates decreased in comparison with the baseline data, but PCR positivity was still detectable. In northern Chiapas and Oaxaca, with no baseline data, there was an absence or near absence of infected and infective flies.

Malaria vectors, *Anopheles hyrcanus* **group (Culicidae, Diptera): morphological comparisons & molecular analysis**

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Anopheles hyrcanus group consists of at least 30 species, with several species including *Anopheles sinensis, lesteri, pullus*, that are known or potential malaria vectors. The morphological diagnostic features and molecular identities of malaria vector *hyrcanus* group species are determined. Larvae, pupae and adults of selected species were described morphologically, and their ITS2 and DNA sequences from our laboratory and GenBank were compared.

Distribution & bionomics of malaria mosquito vectors, *Anopheles (Anopheles)* in Japan, Korea & other parts of Asia

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Mosquito specimens of *Anopheles (Anopheles)* from collections conducted in Japan (2002-03), Korea (1999-2003), Philippines (2002), HongKong (2001), mainland China (2001-03) were identified using morphological and molecular techniques, and recorded. Larval, pupal and adult rearings of field collected samples were conducted for morphological and molecular analysis. Detailed locations of various collection sites and habitats were characterized for known and potential malaria vectors of different countries. Habitats of vectors include ricefields, irrigated ditches, temporary and permanent pools, ponds, streams, others.

Dengue mosquito vectors, *Aedes* and *Ochlerotatus*: morphological identifications & distribution

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The diagnostic morphological features of mosquito vectors of dengue viruses are compared for various *Aedes (Diceromyia, Stegomyia)* and *Ochlerotatus (Finlaya, Gymnometopa)* species found in four regions of the world, namely Oriental Region, Pacific and Australian Region, Afrotropical Region, and the Americas. Morphological characters are indicated to separate both larvae and adults of the vector species of each region. The distribution and bionomics of *Aedes* and *Ochlerotatus* vector species are included.

New & resurgent arboviral disease threatening humans in Australia

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Australia has a range of indigenous arboviruses and an introduced few that are of public health concern; all are mosquito-borne. Endemic encephalitogenic viruses (Murray Valley Encephalitis, Kunjin) are annually active and responsible for occasional cases in the northwest but, despite widespread vector distribution, only rare activity and no epidemics in the southeast since 1974. The most common arboviruses, Ross River annually infecting many thousands nation-wide causing polyarthritis, and Barmah Forest (a similar syndrome) on the increase and responsible for outbreaks of hundreds regionally, have a spectrum of coastal saline and inland freshwater vectors. The most important exotics are the returning dengue viruses with outbreaks of hundreds confined to Queensland by vector distribution but with virus endemicity and vector spread threatening, and the recently introduced Japanese Encephalitis virus causing only a few deaths in northern Queensland but with virus persistence and its affinity for a local vector threatening. Environmental factors drive arbovirus activity by influencing vector populations, which vary with virus and circumstance, although little is known of the role of reservoir hosts. Tourism and development activities that increase human/vector/virus contact, and climate changes that promote vector and virus proliferation, threaten to exacerbate the disease threat to the public health.

Repellents for integrated tsetse management

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ICIPE is in the forefront in identifying potent repellents, both synthetic analogues of natural tsetse repellents and natural repellent blends from un-preferred animals like waterbuck and zebras. One potent synthetic repellent for savannah tsetse species, developed by molecular optimization studies, acts as an olfactory antagonist of a key phenolic kairomone, which flies use to locate the host for feeding (Saini & Hassanali; patent application). This repellent substantially reduce tsetse challenge (> 80%) and feeding efficiency (>80%) on cattle. A prototype dispenser which is placed around the neck of an animal has been developed. Field trials indicate that the repellent can provide effective protection to cattle but all animals in a herd need to be protected or else significantly higher infection occurs in unprotected animals. Experiments are ongoing to evaluate how repellents can be integrated with other existing tsetse control techniques. A 'push-pull' strategy that deploys repellents (to 'push') and baited traps / targets (to 'pull') to enhance suppression rates is being evaluated. A variant of this may involve the use of a proportion of cattle with 'pour on' insecticides as live baits. A business plan for commercial production and distribution of repellent products is also being developed.

Mitochondrial genome organization of lice is highly variable

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Mitochondrial (mt) genome organization is extremely conserved in mammals, vertebrates and most arthropods, e.g., humans and frogs have the same mt genome organization; so have fruit flies and water fleas. We have sequenced the entire mt genomes of the wallaby louse (Phthiraptera: Amblycera) and the pigeon louse (Ischnocera), and part of the mt genomes of the human louse (Anoplura) and four other amblyceran lice. Unlike most arthropods, the mt genome organization of these lice is highly variable. Lice share only 1-2 gene boundaries with fruit flies. No gene boundaries were shared among lice of the three suborders. Mt genome organization differs even among three genera within the suborder Amblycera. Changes in the mt genome organization of lice include gene translocation, inversion, separation of rRNA genes and loss or multiplication of non-coding regions. In addition to lice, we found mt genome organization was variable or highly variable in other two hemipteroid orders, Psocoptera (psocids) and Thysanoptera (thrips), but is strictly conserved in the order Hemiptera (bugs). We speculate that the changes in the mt genome organization in lice, psocids and thrips may be related to their life-history traits such as small body size, small colony size, and high degree of inbreeding.

Forensic entomology in cars

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To determine the entomological succession on a decomposing pig inside a vehicle following its sacrifice by CO poisoning (suicide pig), to calculate the time of decomposition of a suicide pig and determine temperature fluctuations inside vehicles. The significance of this research is the requirement by police and the justice system for accurate information on the Post Mortem Interval (PMI) of cadavers, particularly in regard to homicides and suicides. Most suicides conducted in vehicles happen in isolated areas where the vehicle is parked in wooded areas for the purposes of concealment. Generally this is a shaded area or an area with minimal exposure to the sun. The methodology includes sacrificing 40/45kg pigs using CO gas. Following death the pigs are quickly dressed and placed inside a vehicle. CO gas is pumped into the vehicle for another 5 hours. Decomposition is recorded using infrared video cameras as well as daily observations of insect activity. Decomposition rates are compared with two other pigs (one pig sacrificed by CO poisoning and the other by head bolt) decomposing under normal conditions. Each pig has a datalogger placed inside its throat, abdomen and colon. The temperature inside the car is also measured and compared with the recorded ambient temperature.

The results of a clinical trial of a novel pediculicide

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Few families & medical practitioners in westernised countries are spared the challenge of curing an infection of head lice. Indeed, around 1 in 4 to 1 in 3 primary school children in Australia & other westernised countries may have lice and/or eggs (dead or alive) at any point in time. I report on the first ICH-GCP double-blind randomised controlled trial (RCT) of a pediculicides & the first double-blind RCT of essential-oils pediculicide.

Successful Outreach in Entomology - Packaging the Product

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Successful entomology outreach depends on attracting the consumer to the activity or product. Such marketing is similar to that used by industry to sell ideas and materials. Humor, novel activities, and costumes are effective tools to create interest in entomology.

Assessment of bacteriophage WO involvement in *Wolbachia*-induced CI within the *Culex pipiens* complex

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The *Culex pipiens* complex includes some of the most medically important mosquitoes due to their cosmopolitan distribution and their capacity to vector infectious diseases. Members of the complex differ in their ecology, behavior, physiology and vector competence. To date, no consensus has been achieved about the taxonomical status of representatives of this group. Opinions of their relationship range from distinct species with no gene flow to physiological forms with considerable introgression. Most populations of *Culex pipiens* are infected with *Wolbachia*, a maternally inherited bacterium, which causes cytoplasmic incompatibility (CI) resulting in embryonic mortality. In prior molecular characterization, we have demonstrated that *Culex* populations differ in the bacteriophage WO that is thought to infect *Wolbachia* (Sanogo and Dobson, 2004). To test the hypothesized involvement of bacteriophage WO in determining CI in *Culex*, crosses have been conducted between nine *Culex* strains that differ in their bacteriophage WO infection types. Here, we will present the crossing results and discuss the correlation between bacteriophage WO and CI type. We will also discuss the results in relation to the restriction of gene flow among naturally occurring *Culex pipiens* populations. Reference: Sanogo YO and SL Dobson. 2004. Insect Molecular Biology (*in Press*)

Monitoring the mosquito *Aedes aegypti*: A novel surveillance method and new entomological indices using the gravid trap MosquiTRAP^{â} and a synthetic oviposition attractant (Atr*Aedes*^{â})

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MosquiTRAP^{â} is a novel, simple, easy, low cost, and efficient trap especially developed to catch *Aedes.* mosquitoes. It relies on visual cues and synthetic oviposition attractants (Atr*Aedes*^{â}), based on volatiles identified from grass infusions. Compared to ovitraps, MosquiTRAP^{â} allows the identification of the mosquito species in the field, thus saving time and avoiding laboratory routine such as counting eggs and larval identification. Trapped mosquitoes can also be used for virus diagnosis. New entomological indices are (a) the Positive MosquiTRAP Index (PMI), the percentage of positive traps, and (b) the Adult Density Index for *A. aegypti* and for *A. albopictus*. Field data can be collected using hand-held PDAs and then loaded directly into a Geographical Information System (GIS), for an efficient determination of local entomological indices. At the moment, a national monitoring program in Brazil using this technology is being established.

Tsetse.org and Tsetse Plan: interactive programmes to assist planning control of tsetse.

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"A range of techniques is available for the effective control of tsetse flies - so why are tsetse still a problem over much of Africa? Part of the answer is that control operations have not been planned and executed with sufficient understanding. Non-specialists play an increasingly important role in planning and implementing tsetse control operations, but the diminishing number of specialist entomologists makes gaining expert advice ever more difficult. Handbooks to ensure better use of available technologies exist, but these are no substitute for the specialist advice required to develop a strategy suited to local circumstances. To address this knowledge gap, the interactive programme 'Tsetse Plan' has been developed. The programme is a computerised simulation of the interactive help that would be available if a farmer or NGO was able to talk with a specialist on-site, to assess the local situation, draw up a control strategy, identify technical specifications and prepare a budget. The programme is underpinned by a simulation model of a tsetse population which allows the user to compare the impacts of various control operations. The programme is available via a website (http://www.tsetse.org) which provides further technical advice and slide shows on use of control technologies.
"

An instructional DVD on collecting entomological evidence for court

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This DVD was created by forensic entomologists to fulfill the immediate need to standardize practices for collecting entomological evidence. The material is intended to instruct forensic investigators on how to collect specimen samples and field data in a way that will be scientifically valuable and credible in court. The DVD includes an overview of forensic entomology and the decomposition of an animal model has been time-lapsed to demonstrate the association of insects with various stages of decay. Insects commonly found with human remains are shown and factors that influence insect activity and development are described. Crime scene photographs and video are used to aid in the recognition of what information should be collected during death investigations. A list of equipment needed to process entomological evidence is given, and the collection and preservation of insect specimens has been clearly demonstrated in a step-by-step procedure. The main purpose of this DVD is to standardize the practices which are critical to the development of postmortem intervals and the utilization of insects as evidence during criminal investigations. The entire training video is 45 minutes; however, the multi-media format provides self-paced instruction and allows the viewer select specific modules for quick referencing.

The mosquitoes of Europe - an identification and teaching software

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The bilingual CD-ROM (English/French) proposes identification software, which allows the identification of 100 species of Culicidae (larvae, female and male adults of the genera *Aedes, Anopheles, Culex, Culiseta, Coquillettidia, Ochlerotatus, Orthopodomyia* and *Uranotaenia*) and a knowledge base on these mosquitoes. The study area includes 37 countries of geographical Europe plus Madeira. Contrary to the dichotomy keys, the "identiciel" allows an identification by permanent and intuitive multicriterial approach. With each step, the user selects one of the criteria suggested among morphology, ecology and distribution. He progresses towards the identification of the species by successive choices suggested by images. At the end of the process, taxonomic cards authorise a meticulous checking of the result as well as a comparison with close species. More than 800 original illustrations are provided, the software also gives synthetic information and brings up to date data on taxonomy, bio-ecology, distribution, medical or veterinary importance of these dangerous hematophagous insects, as well as methods of control. An indexed bibliography of 325 references is also included. Easily used, this CD-ROM is intended for teachers, students, medical entomologists, parasitologists, and all agents of human health or veterinary surgeons in charge of monitoring and control of mosquitoes.

Bionomics of *Anopheles hyrcanus* in resurgent and endemic foci of malaria in Iran

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An.hyrcanus group is reported from different parts of Iran. This species plays a role in malaria transmission in neighboring countries. The adult and larvae were collected from different parts of Iran using WHO's methods. This species is found in shaded part of rice-lands and irrigation canals and shaded stagnant water in Nikshahr and Pars-Abad and Poldasht districts. Its peak of activity occurs in midnight whereas s area three peaks of biting can be observed, mainly sunset and midnight according to locality. Light trap is able to collect the adult. This species tend to rest indoor when temperature is falling dawn. The peak of its activity is mainly in August. From the field study it was concluded that *An.hyrcanus* population found in different resurgent and malaria endemic area of Iran. Their seasonal activity, biting habitat, larval density, and abdominal condition is varied in different localities. It seems that in some area due to high population density and antropophilicity of this species it may play a role in malaria transmission. At the future, further molecular and biochemical studies will be done to confirm different sibling species of this species as in studied area.

Behavioral responses of *Culex* to oviposition site odors

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Culex mosquitoes, as important vectors of arboviruses in North America, are effectively sampled by gravid female traps baited with infusions such as hay infusion. Using several methods, a wide range of volatile compounds have been identified from hay and grass infusions and dairy effluent. The purpose of this study was to evaluate the behavioral responses of gravid *Culex* to these compounds as oviposition site attractants in a dual-port olfactometer assay. Responses obtained to individual compounds and then in binary and/or tertiary blends were compared to hay infusion. The efficacy of lures incorporating these mixtures were also evaluated in the olfactometer and compared to hay infusion standards. These results contribute to the development of standardized lures for gravid *Culex*.

Effect of fire on recovery and identification of entomological evidence at a homicide scene

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Insects are valuable in determining elapsed time since death in a homicide. However, the killer may attempt to disguise the crime by setting fire to the crime scene. Experiments were conducted to determine the effect of arson on entomological evidence. Three freshly killed pigs (*Sus scrofa* L.) were placed in separate rooms in a house in Edmonton, Alberta. Extensive blow fly (Diptera: Calliphoridae) colonization occurred, and the remains were allowed to decompose to the advanced decay stage. At the time of the fires, large numbers of larval Calliphoridae, pupae and empty puparia, as well as teneral flies were present. Four successive simulated arson fires were set then extinguished. The first fire was ignited using gasoline poured on the carcass, the second used common household accelerants such as newspaper, the third was set in the basement and the final fire involved the entire house. After each fire was extinguished, the carcasses were examined for entomological evidence. In all, cases, enough entomological evidence was recovered after each fire, including the final destruction of the house, to allow an entomologist to analyze the evidence and to determine elapsed time since death. In several cases, live insects were recovered.

Novel strategies for arbovrius surveillance in response to WNV

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The outbreak of West Nile virus (WNV) in 1999 in Queens, New York USA was an unprecedented public health event on the North American continent. Albeit, the epidemic has stimulated development of only a few novel vector/virus surveillance tools. The VecTest® is the best-known novel technology resulting from the outbreak of West Nile virus. This lateral flow or dipstick test is used to detect West Nile virus antigen in homogenized mosquitoes. The VecTest has also been used successfully to detect viral antigens in extracts of cloacal swabs taken from moribund WNV-infected birds. Another more recent diagnostic test, the RAMP West Nile Virus Test, detects WNV antigen in mosquitoes and birds. This test is based on latex particles that are tagged with fluorescent antibodies to WNV. Sentinel animals have been used to detect the occurrence of WNV. The virus is highly lethal to some birds, such as American crows and blue jays. Field surveillance of WNV activity involves a strategy of systematically collecting and testing dead corvids for WNV infection. Counts of dead corvids have also been used to estimate the intensity of local transmission of WNV.

Weather station ambient temperature corrections in forensic entomology

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Weather station data is used by forensic entomologists to estimate ambient temperatures at body discovery sites. Correlation data from sites may also be collected to correct for site and station differences. This experiment tested the accuracy over time of retrospective weather data correction using linear regression between stations and sites. Temperatures were logged at six hypothetical body discovery sites during a seven day period for which a hypothetical body lay *in situ*, and a further four correlation periods. The accuracy of weather data for hypothetical body *in situ* periods improved after correction in 22 of 24 correlations, however, mean predicted body *in situ* temperatures for sites differed significantly between correlation periods. Predicted temperatures generally rose with time after body removal, which correlated with rising site temperatures accompanying seasonal change. Therefore, practitioners should be cautious in making correlations if weather patterns during correlation differ greatly from those while the body was *in situ*.

Biometrics and behavior in mosquito repellent assays

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Biotic and environmental factors influence the outcome and interpretation of repellent bioassays. Mosquito-related test factors include larval nutrition, carbohydrate availability, age/parity of females, blood engorgement status, and diel blood-feeding patterns. Environmental/human subject-relevant factors are light, temperature, humidity, repellent dose and exposure time, absorption, penetration, evaporation, abrasion, and/or washing of repellent from skin (by perspiration), and the innate repellency of human/animal test subjects. In laboratory tests, cage configuration is critical. With small cage/modular-types of test apparatus (<1000 cm^3), treatment effects may be confounded with "edge effects", because of the position of a feeding port or module, whereas tests repeated using the same human subject comprise pseudo-replication. In large cage (>1000 cm^3) tests, complete protection times are based on the responses of mosquitoes in the upper extreme of the frequency distribution for repellent tolerance, rather than on the average population response; large cage techniques are not useful for determining the Estimated Median Dose (ED$_{50}$), or other percentiles of interest. Variation in repellent assays can be minimized by rearing and testing mosquitoes in a standardized environment, by choosing appropriate sample sizes, by selecting a biologically-relevant assay method, and by careful and diligent oversight of test proceedings.

Vector and viral genetic determinants of epidemic dengue and dengue hemorrhagic fever in Mexico

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To elucidate vector determinants of dengue transmission in Mexico, ~1,600 *Ae. aegypti* collected from throughout North America and individually phenotyped for dengue 2 (DEN2) virus midgut and disseminated infections. DNA from each of these mosquitoes was archived for association mapping of candidate genes that condition vector competence. Dramatic differences in vector competence in the *Aedes aegypti* populations were demonstrated, and vector competence was correlated with dengue seroprevalence. Studies to reveal the genetic basis of *Aedes aegypti* vector competence identified two candidate genes (Early Trypsin and Abundant Trypsin) that condition dengue virus infection of vectors. RNAi silencing of the Early Trypsin gene resulted in reduced vector competence of *Aedes aegypti*. To determine the epidemiological relevance of these genes, a population genomics approach using association mapping was used to reveal the statistical relationship between genetic polymorphisms and vector competence phenotype in these genes. An association was found between the *Abundant Trypsin* and DEN2 disseminated infection rates DEN2 infection phenotypes in these American *Aedes aegypti* populations. Such information is important in risk assessment for epidemic dengue and DHF/DSS. During these studies, a new DEN2 virus of the American-Asian genotype was introduced in Yucatan State, causing an epidemic of DHF/DSS. This virus is being compared with other DEN2 viruses, which have previously circulated in the Yucatan State, for its ability to infect, replicate in, and be transmitted by *Aedes aegypti* and for its epidemic potential.

Chemical analysis and laboratory bioassays of host odors to identify compounds that attract and inhibit the attraction of mosquitoes

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Using various sampling techniques and gas chromatography/mass spectrometry (GC/MS), a comprehensive chemical profile of host odors can be obtained. The differences in chemicals emanated by different hosts may provide a clue to the structures of compounds used by anthropophilic mosquitoes, compared to those used by zoophilic mosquitoes, for host location. Screening of candidate compounds is accomplished in our laboratory by using a triple-cage dual-port olfactometer. Bioassays of single compounds and blends of compounds have resulted in the development of attractive synergistic binary blends for anthropophilic mosquitoes. Further improvement to the blends has resulted in a three-component blend that is more attractive than some human individuals in laboratory bioassays. During the development of these attractive odor blends, some compounds were observed to inhibit the host-seeking capability of mosquitoes, possibly my masking the attractive odors. Inclusion of these compounds (attraction-inhibitors) in attractive blends, results in a substantial decrease in the ability of mosquitoes to locate the attractive odor source in the olfactometer. Laboratory bioassays are being used to develop a system that demonstrates the possibility of concealing a host with attraction-inhibitors and driving mosquitoes to choose an attractant blend over a live human or animal host.

Sexual dimorphism of antennal gene expression in the malaria vector *Anopheles gambiae*

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To obtain a better understanding of the olfactory processes that allow mosquitoes to identify human hosts, a molecular study has been performed to identify and characterize molecules in the olfactory signaling pathway of the African malaria vector *Anopheles gambiae*. cDNA libraries from antennae of females and males were established, from which a collection of cDNAs encoding odorant binding proteins (OBPs) and other novel antennal proteins were isolated and characterized, representing various families of putative carrier proteins with homologues in other insects. Using filter array hybridizations and quantitative RT PCR, regulation and gender specificity of expression of these genes was investigated. Although male and females appear to express the same olfactory genes, significant differences in steady state levels of some of these putative carrier protein genes were detected between the sexes and after blood feeding in females. Results of microarray experiments of gender-specific expression of olfactory carrier proteins and receptors will be reported. Our ultimate goal is to identify and characterize molecules in the odor reception pathway of this mosquito vector species with the intent of facilitating the development of better methods to interfere with the mosquito-host interaction.

Study of decomposition rates of pig carcasses and viability of *Calliphora vicina* in the UK climate

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The estimation of post mortem interval (PMI) in a country such as the UK which experiences fluctuating temperatures, both seasonally and daily, is considerably more complicated than in those countries with fairly constant temperatures. In order to assess the effects of the different seasons on decomposition rates and insect species composition, piglet carcasses were put out at The Natural History Museum in Central London. Significant differences were recorded from carcasses put out at different times of the year, and between those laid outside on a roof, and those inside the building, where insects had limited access to the carcasses. Although oviposition by the primary invaders occurred at the same time on both the indoor and outdoor carcasses, the rate of development of *Calliphora vicina* on the outdoor carcasses was significantly slowed down due to the cold temperatures experienced, thus limiting the decomposition rate considerably. Further laboratory based studies were carried out to assess the effect of cold temperatures on the different life stages of *Calliphora vicina*.

Molecular basis of resistance to pediculicides

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Head and body lice develop resistance to pediculicides through mutations of insecticide target sites, such as the voltage-gated sodium channel, and increased activity of detoxification enzymes such as oxidases and esterases. The difficulties of conducting bioassays on field-collected lice make molecular analysis of insecticide resistance an especially attractive option. Here are described the molecular details of resistance that has appeared in lice together with a discussion of ongoing work to develop methods to detect and assess these mechanisms through molecular analysis. A useful approach to measuring all of these mechanisms is fluorometric PCR. This method allows detection of resistance mechanisms based on point mutations at the insecticide target site, resistance gene amplification, or increased expression of resistance genes. Knowledge of resistance gene sequence is a prerequisite in the development of these methods. Our knowledge to date of louse resistance gene sequences is summarized. The same molecular methods that serve to detect resistance are also central to the determination of population genetic relationships of the resistance alleles. We discuss our present state of knowledge of louse molecular markers and the louse genome as they relate to insecticide resistance.

Environmental change and lyme disease risk

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An understanding of the influence of environmental change on Lyme disease in North America is a fundamental component in assessing changes the spatial distribution of human risk. Because disease risk may be influenced both by large-scale climate change and local-scale landscape modification, we developed a multi-scale approach to dissecting the influence of environment. First, we used a climate suitability model for the tick vector *Ixodes scapularis* to examine the potential effects of global climate change on future Lyme disease risk in North America. The model reveals a significant expansion of *I. scapularis* north into Canada and central United States as well as a retraction of the vector from southern United States. With higher resolution remotely sensed data, we evaluated the impact of local-scale landscape structure on both entomologic risk and human exposure. Landscape fragmentation resulting from suburban development was compared to both human and tick surveillance data from Connecticut and showed that landscape mosaic plays a significant role in defining disease risk. In spite of higher tick densities, human incidence of Lyme disease was found to be lower in fragmented contexts. The synthesis of such environmental models will allow for improved predictions of Lyme disease emergence.

Influence of the structure of host-odor plumes on the orientation maneuvers of flying mosquitoes

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Host finding in mosquitoes is typically subdivided into two sequential stages: attraction at long range that is induced by host-released odors, principally assumed to be carbon dioxide. Orientation at close range is assumed to be regulated by the interactions of carbon dioxide with other host odors, humidity, heat, and visual cues. In our work with *Aedes aegypti* in a wind tunnel, upwind fight along the carbon dioxide plume required the sequential encounter with filaments of carbon dioxide, as would normally occur in a plume diffusing in wind. A single brief encounter with a filament of carbon dioxide odor induced a surge toward upwind. A relatively homogeneous cloud of carbon dioxide, in contrast, activated flight, but did not stimulate upwind movement. Conversely, orientation to human skin odor seems to require a longer term encounter than occurs with exposure to an odor filament. The threshold for response to skin odor is itself modulated by an encounter with carbon dioxide: a brief exposure to carbon dioxide lowers the threshold of response to skin odor by five-fold. Together the interactions of carbon dioxide and skin odor explain several features of host orientation, and provide support for the hypothesis of sequential stages of odor utilization.

Development of lure for the dengue mosquito

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Instead of proceeding with the traditional isolation and identification from hosts of individual mosquito attractants, ISCA Technologies, Inc. created compositions which mimic the skin of the specific vertebrate hosts. Most mosquitoes apparently exhibit some level of host 'preference', i.e., they do not feed indiscriminately on any vertebrate host. In some studies host-feeding patterns within a closely related species complex is odor mediated and was attributed to differential response to CO_2 and other skin odors. The actual bait, created through a process patented by ISCA, provides the parasitic arthropod (host seeking *Aedes aegypti*) a much more complete "olfactory sensation" associated to a specific "host" (human) than any other attractants on the market. Traps lured with this bait containing artificial "sensory correct" profiles of human hosts trigger rapid responses from females *Ae aegypti* inducing the full set of host seeking and feeding behaviors, from activation to imbibing. The current ISCA prototype trap has proven to be superior to the Fay-Prince and Wilton traps and other commercial traps set-up in competitive laboratory trap tests. Laboratory studies indicate the capture rate of this prototype baited with this new lure closely approximates the rates obtained from human biting counts for the same mosquito density.

2-Oxocarboxylic acids elicit landing responses from female *Anopheles gambiae*

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Landing and probing responses were elicited from female *Anopheles gambiae* in wind tunnel bioassays by 2-oxocarboxylic acids. The most significant responses were elicited by 2-oxopentanoic acid although 2-oxo-butanoic, 2-oxo-3-methylbutanoic, 2-oxo-3-methylpentanoic, 2-oxo-4-methylpentanoic and 2-oxo-hexanoic acids also elicited responses. 2-Oxo-propanoic (pyruvic), 2-oxo-octanoic, 2-hydroxy-propanoic (lactic) and 2-hydroxy-pentanoic acids were inactive. The response to 2-oxopentanoic acid was dose and temperature dependent with 30 µg and body temperature (34^{o}C) eliciting the most significant responses. Responses decreased over time but this was thought to be due to evaporation. Chemical analysis of a suitably derivatized human sweat sample confirmed the presence of pyruvic, 2-oxo-3-methylbutanoic, 2-oxo-3-methylpentanoic and (*E*)-oxo-4-methylpentanoic acids. The three methyl-branched acids, previously identified in human body fluids, are metabolically linked to the amino acids valine, leucine and isoleucine *via* a reversible trans-amination. However, the most active compounds, 2-oxopentanoic and 2-oxohexanoic acid were not identified in human sweat. Taken together the research suggests that C_4-C_6 2-oxocarboxylic acids represent a class of compounds that provide female *An. gambiae* with close range, host-specific odour cues.

Distribution of blood feeding within the subgenus *Inseliellum* (Diptera: Simuliidae)

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Simulium (*Inseliellum*) *buissoni* in Nuku Hiva, Marquesas Islands, Polynesia, has been referred to as the "scourge of the Pacific". It is a sufficiently serious blood feeder on humans that it resulted in the administrative centre of the Marquesas being moved from Nuku Hiva, south to Hiva Oa. *Simulium buissoni* is also a serious bird biter and attempts to eradicate this black fly were made in the early 1990's. *Simulium* (*I.*) *gallinum,* distributed on other Marquesas Islands, however, feeds only on birds. Distribution of blood feeding within *Inseliellum* will be discussed in light of a reconstructed phylogeny for the subgenus. Also discussed will be the "Miracle of the Nono" from the 1930's, where it is reputed that serious biting of humans by black flies on the southern Marquesas Islands, ceased ostensibly, overnight.

Evolution of blood-feeding behavior in black flies (Diptera: Simuliidae)

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Phylogenetic-, paleontological-, and host specificity data are combined to reveal patterns of blood feeding behavior among black flies. The fossil record reveals that simuliids originated before the radiation of birds and mammals, suggesting that other warm-blooded hosts were exploited by the earliest members of the family. Ornithophily probably evolved independently in the Prosimuliini and Simuliini, with numerous, independent, reversals to mammalophily in the latter tribe. Although host-switching events in black flies have been predominantly from ornithophily to mammalophily, there is a least one example of the reverse among certain members of the *Simulium* (*s.str.*) *slossonae* species-group. Obligatory autogeny (i.e., females that lack biting mouth parts), is a derived trait that evolved numerous times independently from both mammalophilic and ornithophilic progenitors. Such non-bloodsucking species, which constitute only about 2.5% of the world species, live predominantly in high latitude or high altitude environments. Tarsal claw morphology has utility for inferring host preference among anautogenous black flies; however, the current practice of coding claws as either "simple" or "bifid" does not exploit the full panoply of variation present in this character system. Detailed comparative studies are needed in order to identify useful characters for species recognition and phylogenetic studies.

Landscape epidemiology of malaria in Tumkur (Karnataka), India, as monitored by satellite remote sensing

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Study was conducted in selected villages of 3 Primary Health Centres of Tumkur (Karnataka)with varying endemicity to find out the landscape elements critical to malaria endemicity at village level using satellite remote sensing.. Ground data on the presence of breeding habitats, monitoring of larval density, adult *Anopheles culicifacies* (malaria vector) from indoor resting sites and ecological features within 1.5 km radius from the centre of each village were generated in 2001 and 2002. Data of Indian Remote Sensing satellite with 23.8 metre and 5.8 metre resolution respectively were procured for the dates of ground survey and analyzed for generation of images and statistics of land use features within a buffer zone of 1.5 km around each village. Delineation of tanks, streams, ponds, marshy areas, the main breeding habitats of *An. culicifacies* was possible by Remote sensing by merging LISS III and PAN products. The breeding habitats in the vicinity of human settlements within 500 metres were found more productive for *An. culicifacies.*

Quantification of land use features revealed that extent of water bodies, permanent vegetation cover, less barren/scrub and rocky areas were associated with high malaria endemicity. Identification of villages into high/low risk of malaria is possible leading to village level stratification for development of village specific selective and cost effective spraying strategy for integrated vector management programme for targeting malaria control.

Aedes aegypti **behavior in manipulated domestic environments in Thailand**

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Aedes aegypti, a low-density mosquito, lives in or near human dwellings, feeds frequently and specifically on people, and efficiently transmits dengue viruses. Field experiments with wild mosquitoes are risky in endemic areas and population augmentation to increase sample sizes causes ethical concerns. We circumvented both issues by screening the entire house and yard of rented homes in order to safely conduct relatively natural behavioral experiments using females collected in the village as pupae. Host size, location, and orientation effects on feeding were studied using 4 rotated subjects. Released mosquitoes (~25%) were recovered using pack-pack aspirators and the origin of their blood meals scored by DNA fingerprinting. There was strong correlation between host size and feeding with the largest individuals consistently feeding the most mosquitoes. *Ae. aegypti* fed most often on people inside house but body orientation appeared unimportant. Timing of oviposition and the impact of container size on egg number were evaluated in various sized jars lined with germination paper. Papers were changed at regular intervals and eggs counted. Most eggs were laid in late afternoon. Larger diameter containers consistently received more eggs whether located inside or outside. Indoor females preferred containers without preexisting eggs.

Behavioural responses of gravid *Aedes aegypti* (Diptera: Culicidae) to synthetic oviposition attractants identified from grass infusions volatiles.

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Grass infusions have been used as oviposition attractants and/or stimulants for enhancing ovitrap efficacy. However, the infusion procedures are very time-consuming and extremely labour-intensive. Thus, the volatiles released from grass infusions that attract gravid mosquitoes *Aedes aegypti* to ovitrap were identified by coupled gas chromatography-electroantennogram detection (GC-EAD), coupled gas chromatography-masspectometry (GC-MS), electroantennography (EAG) and field trapping. Six candidate compounds were identified and evaluated in the field and in laboratory. Electrophysiological studies showed that antennas of gravid mosquitoes are sensitive to all synthetic compounds but only few individual compounds elicited behavioral responses within a dual port olfactometer. Laboratory and field studies showed that the lures and dispensers are still attractive after 30 days old in the environment. Video recording analysis of the behavior response of gravid *A. aegypti* to adult mosquito trap (MosquiTRAP^{â}) baited with the oviposition lure (Atr*Aedes*^{â}) showed that chemical and physical structures of odor plumes have great influence on the trap catch performance. At the moment, the MosquiTRAP^{â} and the Atr*Aedes*^{â} are being evaluated for *A. aegypti* national monitoring program in Brazil.

Laboratory and field evaluation of mosquito repellents in Australia

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This paper will review laboratory and field studies conducted in Australia and Papua New Guinea to evaluate mosquito repellent formulations. The response of *Anopheles* spp. in the field to *N,N*-diethyl-*3*-methylbenzamide (deet) is variable, and several species are tolerant of current formulations. In contrast, deet repellents are quite effective in providing protection against Culicine mosquitoes in this region. Additional studies on the laboratory and field response of mosquitoes to a comparatively new active ingredient, (1-methyl-propyl 2-(2-hydroxyethyl)-1-piperidinecarboxylate), known as picaridin, will also be presented.

Role of defensin in tick-pathogen interaction

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Antimicrobial peptides are major components of host innate immunity, a well-conserved, evolutionarily ancient defensive mechanism. Infectious disease-bearing vector ticks are thought to possess specific defense molecule that have been acquired in the process of their evolution against the transmitted pathogens. We found a parasiticidal peptide evolved from a common ancestral peptide resembling spider and scorpion toxins from *Haemaphysalis longicornis*, the obligatory vector for *Babesia* parasite transmission. This protein, named longicin, displayed bactericidal and fungicidal properties resembling those of defensin homlogues from invertebrates and vertebrates. Longicin showed a remarkable ability to inhibit the proliferation of merozoites, an erythrocyte-blood stage of equine *B. equi*, by killing the parasites. Longicin was localized at the surface of the parasites, as demonstrated by confocal microscopic analysis. In an *in vivo* experiment, longicin induced significant inhibition of parasitemia in animals infected with the zoonotic and murine *B. microti*. Our findings suggest that longicin is a promising candidate for designing antibabesia therapeutics.

Attractive host odours for mosquitoes: the blend ratio makes the difference

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Mosquito species differ with respect to their host preferences, and some discern not only different species of vertebrate hosts but also different individuals of a given species. Olfactory cues derived from the hosts determine to a large extend the host choice, and it is an intriguing question how these insects accomplish the necessary fine odour discrimination. Emanations from vertebrate hosts consist of complex blends. Attention has focused on the identification of kairomones that guide mosquitoes to their hosts. Here we show that odour blends rather than their single constituents attract the anthropophilic yellow fever mosquito *Aedes aegypti* (L.). Highly attractive blends consist of L-(+)-lactic acid, ammonia, and distinct fatty acids which were essential to maximise the attractiveness of the blend. The mosquitoes' responses, however, depended markedly on the ratio of these compounds. In a direct competition bioassay a distinct blend ratio turned out to be as attractive as an average human hand. When added to natural human skin emanations some compounds also altered significantly the attractiveness of the host. The data show that the blend ratio of a few distinct compounds is crucial for the attraction of mosquitoes to human hosts.

Specific monitoring tools for anthropohilic mosquitoes

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For a better estimation of the risks of mosquito-transmitted disease epidemics, it is vital to quickly assess the density of the host seeking mosquito population. A simple, specific, and effective trapping tool would overcome some of the disadvantages of the currently used methods to estimate the density of mosquito populations. We introduce an new monitoring suction trap with an attractive lure for anthropohilic mosquitoes. The lure releases a cocktail of volatiles that also emanate from human skin. The attractiveness of the mixture strongly depends on the mixing ratio of the compounds. Release rates and blend ratio can be optimised to suit a number of anthropophilic mosquitoes. Although CO_2 is not essentially necessary for the effectiveness of the new attractant, it enhances the catching rate of the blend.

The design of the trap combines the generation of a specific plume shape for an optimum effectiveness of the attractants with visual cues to enhance the catch of the attracted mosquitoes. The trap is optimised for the new attractive blend, easy to produce and simple to use. It can be applied indoors as well as outdoors and is currently tested in the laboratory and in the field in Brazil, Asia, Australia and Europe with a range of mosquito species.

Forensic entomology and artificial intelligence: which is the relation?

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Blowflies of genus *Chrysomya* are so important in forensic entomology to indicate the Post Mortem Interval (PMI). Moreover, the larvae of these insects have an aggregation behaviour when are feeding of the tissues in human and animal corpses which each one influence the other one during this process of feeding and post- feeding, and still after, during the post- feeding larval dispersal searching for pupation sites. The results of this influence is a self-organization in this process which is studied in computer programs (Cellular Automata) using the data from this behaviour and the program produced from these data to be applied in the development of programs of Artificial Intelligence and improve the performance of navigation systems in robots by the environment. These studies were done with about 10,000 larvae of *Chysomya megacephala* and *Chrysomya albiceps* in arenas simulating the environment conditions. Therefore, the study of some biological systems just as the studies in Forensic Entomology could be so important to develop programs in Artificial Intelligence.

Role of tick lectins in pathogen transmission

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Tissue specific lectin/haemagglutinin activities of both soft and hard ticks have been investigated. Some tick lectins are proteins with binding affinity for sialic acid, various derivatives of hexosamines, and different glycoconjugates. Most tick lectin/haemagglutinin activities are blood meal enhanced, and could serve as molecular factors of self/non-self recognition in defence reactions against bacteria or fungi, as well as in pathogen/parasite transmission. Dorin M, the plasma lectin of *Ornithodoros moubata*, **is the first tick lectin purified so far from tick haemolymph, and the first that has been fully characterized. Partial characterization of other tick lectins/haemagglutinins has been performed mainly with respect to their carbohydrate binding specificities and immunochemical features. Few papers have been published on tick lectins so far, and therefore more data is needed to complete the mosaic of knowledge of their structural and functional properties in terms of their role in pathogen transmission. (Supported by the Grant Agency of the Czech Republic No. 206/03/1323.)**

A three dimensional pharmacophore model for discovery of new insect repellent compounds

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In pursuit for a better understanding of insect repellent property of molecules we have generated a three-dimensional pharmacophore model using 3D-Catalyst from a training set of eleven diverse insect repellent compounds including DEET. Protection time data for repellent activity of the training set were taken from an earlier published study. Three hydrophobic sites and a hydrogen-bond acceptor site in the molecule appear to be critical for potent insect repellent activity. Calculated stereoelectronic properties such as molecular electrostatic potentials on DEET and some of these molecules in the training set are consistent with our observation. The pharmacophore shows a fairly good correlation (correlation=0.9) between protection time afforded by the compounds in the training set and their predicted protection time. The validity of the pharmacophore model goes beyond the list in the training set and is found to map well on a variety of other insect repellents. By mapping this model on a potent insect repellent compound we have generated a three-dimensional shape based template. A search of in-house database using the pharmacophore template resulted in the discovery of new potential candidate repellent compounds.

Making the case: frequently-contested aspects of forensic entomology evidence

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As forensic entomology evidence is introduced at trial, there are a few predictable aspects that frequently are at the heart of controversy regarding either admissibility or weight. Essentially all of these relate to the argument that, whereas forensic entomology is "good science" generally and applied in the present instance by a qualified expert, that as applied to the facts of the case at bar, the opinion under consideration is not based on reliable data (standards of the U.S. Federal Rules of Evidence or various state codifications thereof). Of those objections related to such "reliable data," the most common related to forensic entomology center around the use of remote temperatures. Few homicide victims are located proximate to weather stations, and fewer still decompose while investigators make measurements. Consequently, in virtually all instances, it is necessary to analyze the thermal development of insect species in the context of temperature data obtained during routine operations of weather stations that typically are located at some distance remote from the site where the decedent was found. Whether these data are analyzed as accumulated degree-days (ADD) or degree-hours (ADH), the same objections are continually raised in court: that the data do not accurately reflect conditions at the decomposition scene (because of uncertainty due to distance, differences in altitude, eccentricities of air flow, cooling effects of adjacent water bodies, heating effect of bright sunlight, or similar concerns). Generally, such objections can be forestalled by simple temperature measurements verifying concordance between the weather stations and body site. Related weaknesses include arithmetic errors in tabulating thermal units, misapplication of ADD to calculate ADH, failure to understand how ADD/ADH are applied in the context of existing dipteran developmental data, failure to consider base temperatures below which development essentially ceases, failure to consider the possible effects of "maggot mass," or putative failure to consider the effect of unique drying conditions on corpses. The forensic entomologist should analyze cases in the context of such potential arguments and be prepared to defend conclusions. Curiously, allegations regarding possible misidentification of insect species are seldom raised.

Blood feeding behavior of west nile virus mosquito vectors in Northeastern USA

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Control and prevention strategies for West Nile virus (WNV) can only be optimized once the most important human vectors have been identified. Although enzootic vectors of WNV have been identified, the epizootic human and horse-feeding vectors are not clear. A three-year study was conducted to investigate the host-feeding patterns of mosquitoes in areas where humans are active and avian reservoirs are nearby. Resting collections, using specially designed vegetation aspirators, were conducted in Upstate New York and New York City. After identification to species, engorged abdomens were analyzed using antibody sandwich ELISA to identify host blood sources. Blood meals also were tested with the precipitin method against antisera for 20 different species of mammals, birds, and reptiles. More than one host in a blood meal was detected in a small number of samples. These results provide a picture of the temporal and geographic variation in feeding patterns of mosquito species commonly included in surveillance programs as well as those that have not been considered important vectors of pathogens in the Northeast USA. The greatest frequency of human blood feeding was found in *Ochlerotatus triseriatus, Oc. aurifer, Aedes cinerius, Oc. fitchii/stimulans, Culex restuans, Cx. salinarius, Cx. territans, Ae. albopictus* and *Oc. japonicus.*

Going global: Taking molecular forensic entomology the next step

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In recent years, the importance of DNA-based techniques for application to the identification of insects, particularly blowfly larvae (Calliphoridae: Diptera) has been widely established. Research has frequently been locality specific, considering the distinction of different species within isolated populations or groups of populations within a small area. While small scale studies are beneficial in development of protocols for accurate molecular identification of insects in specific localities, a global approach to molecular study of forensically significant species facilitates consideration of such issues as species status. This study considers genetic variation in the cytochrome oxidase I (COI) encoding region of mitochondrial DNA (mtDNA) from calliphorids collected throughout Southern Africa and Australia. Alternate regions of DNA are explored for their value in providing an insight to intraspecific variation within several cosmopolitan species. Flies from New Zealand, the United States, Thailand, Taiwan, Brazil and Italy are considered, in addition to Australian and southern African individuals. The study indicates the potential of DNA to provide not only a means of more efficient and accurate identification of insects, but also to analyse the genetic processes occurring within some species, and potential applications and pitfalls for forensic entomology.

The potential for traditional plant-based insect repellents & fumigants as a means of reducing malaria in remote areas

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Insect repellents and mosquito coils are widely accepted as beneficial for travelers in regions of vector-borne disease transmission. However, their efficacy at preventing malaria has never been established and is one reason why such methods have never been extended into disease control programmes. Isolated communities in remote regions are often at increased risk of malaria due to problems in provision of even basic malaria control programmes. For example, *P. falciparum* cases in China are increasing dramatically among minority peoples living in the border regions of Laos, Myanmar and Vietnam, or among migrant forest workers in the Bolivian Amazon. In such areas, particularly where there is an abundant floral habitat such as the SE Asia rainforests or the Amazon, there often remains dwindling historical knowledge of traditional plant-based anti-mosquito measures. By working with such communities it may be possible to identify and develop a means of disease control at the individual, household or village level. Such methods, if effective, could be both cheap and sustainable if local plant species are identified as having repellent, fumigant or insecticidal properties which could be exploited. Surveys of plant use among 25 minority peoples in China are reported and the potential for malaria control assessed.

Incidence, prevalence, and seasonality of *Choanotaenia* and *Raillietina* poultry tapeworms in arthropod and vertebrate hosts

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Confined animal production facilitates arthropod transmission of disease agents by increasing host-vector interactions, fostering sustained exposures, and providing optimal habitat. Poultry tapeworms, *Choanotaenia infundibulum* and *Raillietina cesticillus*, infest domestic and wild fowl throughout the world. Research at the University of California demonstrated that two insect species collected from caged laying hen operations, the hide beetle, *Dermestes maculatus*, and the house fly, *Musca domestica*, were naturally infected with tapeworm cysticercoids. Tapeworm incidence and prevalence fluctuated independently of host populations, with no distinct seasonal pattern. Extrinsic incubation was shown to be temperature-dependent, with development to infective cysticercoids requiring only 5 days at 33C. As husbandry practices change to optimize facilities for food animal productivity, conditions conducive for arthropod populations simultaneously develop. As intensive production increases, infestation rates can be expected to climb as well.

Issues in the control of sheep lice (*Bovicola ovis*)

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Sheep lice are among the three main parasitic diseases of sheep in Australia. Historically sheep lice have been controlled by government regulation and the application of chemical insecticides. However, where regulatory powers still exist they are now seldom exercised and there is increasing sensitivity to the use of chemicals in major wool markets, amongst woolgrowers and in society at large. Woolgrowers face continuing cost of production pressures and the effectiveness of some widely used products has been compromised by the development of resistance. These constraints necessitate a more integrated approach to control. Barriers to change include poor knowledge of the biology of *B. ovis*, difficulty in detecting lice in the early stages of an infestation, lack of availability of alternative control methods and sociological factors. These issues are explored and parallels awn with human head lice control.

Regulation of head lice treatments in Australia

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Head lice treatments in Australia are regulated by the Therapeutic Goods Administration (TGA) via a two-tiered system. Head lice treatments containing herbal ingredients identified as "low risk" (e.g. melaleuca oil) require **listing** on the Australian Register of Therapeutic Goods (ARTG) These products do not undergo efficacy evaluation prior to listing, although sponsors are required to hold evidence to support label claims. Treatments making "high" level claims, or containing ingredients such as malathion, permethrin or pyrethrins require **registration** on the ARTG, involving evaluation of safety, efficacy and quality of the goods. However, a number of currently registered products were available before the onset of the listing/registration process in 1991. Products available before this time may not have been evaluated, although the potential for the TGA to request evaluation of these "grandfathered" products exists. The regulation of head lice treatments in Australia was reviewed in 2002/2003. A number of recommendations resulted from the review including specific recommendations about label claims and the level of evidence required to support registration or listing of a new head lice treatment product. The recommendations take into account the practical requirements of sponsors and the limitations of the current two-tiered regulation system.

A new device for recording host-seeking rhythms of mosquitoes using a photoelectric sensor

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A new automatic device capable of recording the host-seeking behavior of mosquitoes was designed using a photoelectric sensor. The host-seeking rhythms of several mosquito species were recorded in the laboratory. The use of CO_2, in addition to heating and contrast of black and white color, was essential to the functioning of the present recording device. The diel rhythms of non-blood fed *Aedes albopictus*, *Ae. aegypti*, *Culex quinquefasciatus*, *Cx. tritaeniorhynchus*, and *Anopheles stephensi* females recorded by the device corresponded closely with the flight activity levels, as measured by human landing counts and light traps, observed in the field for the same species. This simple automatic recording device also proved to be useful for evaluating the natural host seeking behavior of field-collected mosquitoes.

Astounding success in controlling *Aedes aegypti* in Vietnam using a new strategy

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From 1989, Vu Sinh Nam and Brian Kay have worked together with a team of collaborators including John Aaskov and Truong Uyen Ninh to deliver community based *Aedes aegypti* control using predacious copepods, the programs being managed by the Australian Foundation of Peoples of Asia and the Pacific. The four elements of the new strategy are (1) vertical and horizontal approaches to build up community understanding, (2) quantitative sampling methodologies designed to prioritize control, (3) inoculation of locally acquired *Mesocyclops* into large water storage containers, and (4) vector control activities at community level. By 2003, *Aedes aegypti* has been eliminated from 42 of 46 communes in north and central Vietnam, involving the protection of some 400,000 people. This is a world first achievement as no insecticide was used. The implications of this model are exciting as this approach is applicable to many countries where water has to be stored for survival.

Seasonal studies on the influence of clothing and wrapping on carcass decomposition and arthropod succession

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Six pig (*Sus scrofa* L.) carcasses, three wrapped and clothed carcass and three wrapped and naked, were sampled i) daily ii) after five days and iii) after ten days. Two more carcasses, one naked and not wrapped, and one clothed but not wrapped were sampled daily as controls. Differences in decomposition between the clothed or wrapped carcasses included the distribution and size of maggot masses and significant delay in drying out of the carcasses for all seasons. The maggot masses were primarily dominated by *Chrysomya marginalis* (Wiedemann) and *C. albiceps* (Weidemann), in autumn and summer. *Chrysomya chloropyga* (Weidemann) and *Calliphora vicina* Robineau-Devoidy in winter, with *C. chloropyga* and *C. albiceps* in spring. There was no delay in oviposition except in winter where a five day delay occurred on the wrapped carcasses. Extremely high internal temperatures in the warmer seasons caused significant maggot mortality on the wrapped carcasses. The predation of *C. marginalis* maggots by *C. albiceps* maggots was observed in autumn. In summer, maggot masses of these species were observed in separate groups. The coleopteran community was dominated by *Thanathopilus micans* L. larvae, succeeded by *Dermestes maculatus* De Geer, larvae. Adult *D. maculatus* and *Necrobia rufipes* De Geer, were present on the carcasses throughout all seasons.

Ectoparasites and arthropod-borne parasites of brown rat, *Rattus norvegicus*, in Ahvaz, southwest Iran

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Arthropods play an important role in the transmission of infectious agents to human and animals, such as rodents. Ahvaz, southwest Iran, is encountered with high density of rodents, especially *R. norvegicus*. In this study, 72 *R.norvegicus* were captured from different sites of Ahvaz, using live traps. Their ectoparasites were collected and identified. Thin and thick blood smears were prepared. After dissection of rats, their different organs were examined for any endoparasites, applying specific techniques. In general ectoparasites comprised 2 species of flea, *Xenopsylla astia* and *X. nubica*, 1 species of louse, *Polyplax spinula* and 1 species of mite *Laelaps nuttalli*. Among endoparasites, 9 species utilize arthropods as vector or intermediate host in their indirect life cycles. These included 1 species of protozoan parasite, *Trypanosoma lewisi* and 8 species of helminth parasites. The potential risk of each of these identified species to the health of human and animals will be discussed.

Applications of GIS and remote sensing for surveillance and control of mosquito borne diseases

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The risk of transmission of mosquito borne diseases depends on the spatial distribution of vector species in relation to the distribution of human hosts and, in case of mosquito-borne zoonoses, the distribution of vertebrate reservoir hosts. These distributions and temporal changes in these spatial patterns depend on environmental and climate/weather conditions. Satellite imagery provides means to gather vast amount of landscape/landcover and meteorological data that are proximate determinants of vector distribution and risk of disease transmission. Geographical information systems allow for the integration of environmental, entomological, epidemiological and demographic data and provide a framework for surveillance and control on scales ranging from local health departments and mosquito control districts to national and international programs. Products of applications of these tools and quantitative analysis of spatial data include data management tools, disease risk maps, and decision making tools for location and time of interventions. Examples from studies in the U.S. Israel, Trinidad and Kenya will be presented, including malaria cases in relation to *Anopheles* larval sites in Trinidad and Israel, dengue and dengue haemorrhagic fever in Trinidad, eastern equine encephalitis outbreaks in Michigan, USA, and La Crosse encephalitis transmission and the effect of introduction of *Aedes albopictus* in Illinois, USA.

Suppression of dengue transmission by application of integrated vector control measures at sero-positive GIS-based foci: Demonstration of a cost-effective model

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A serological survey of approximately 2,000 primary school children from six schools in Chachoengsao, Thailand, was performed at the end of the peak dengue transmission period. GIS analysis of sero-positive cases was carried out to determine transmission foci in high IgG-IgM positive villages. Vector control implementation was conducted in the foci and also within 100 meters around the foci, including schools attended by children in the treated areas, with collaboration from local public health and school authorities. Vector control measures included source reduction by routine garbage pickup organized by local government, the use of locally made screen covers, a combination of local Bti and *Mesocyclops*, and locally made lethal ovitraps. Implementation of vector control measures in the foci was continued until the end of the rainy season. Vector control efficiency was monitored using both entomological and serological parameters. Results showed a significant reduction of dengue vectors in treated areas when compared to untreated areas. Despite overall higher dengue incidences in the second year when compared to the first year, IgG-IgM positive students in the treated areas decreased from 13.46% to 0% while those from untreated areas increased from 9.43% to 19.15%.

Novel Mosquito Traps: Do They Really Work

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A diversity of commercial traps for mosquito control have become available during the past five years. Most are marketed to homeowners. Many models have been evaluated in a large outdoor screened enclosure against two species (*Aedes aegypti* and *Ochlerotatus taeniorhynchus*) of laboratory reared mosquitoes to determine their relative efficacy. Several models were selected for field testing against natural populations. Ongoing research is trying to answer the question: Can mosquitoes be trapped out? Different management strategies are being evaluated, e.g. control potential on isolated islands versus urban/suburban residential settings. In urban/residential settings traps are being evaluated as a protective barrier around a city block and in individual yards. Preliminary results indicate that protective trap barriers provide excellent control (>90%) on isolated islands, but only moderate (50% reduction) control in urban/suburban areas. A single trap in a single yard provided no measurable mosquito control.

The mode of action of deet, SS220, and Bayrepel® arthropod repellents against *Aedes aegypti*, *Anopheles stephensi*, and *Phlebotomus papatasi*

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Laboratory bioassays were conducted with human subjects using six-celled K&D modules and the known arthropod repellents, N,N-diethyl-3-methylbenzamide (Deet), (1S, 2'S)-methylpiperidinyl-3-cyclohexene-1-carboxamide (SS220) and 2-(2-hydroxyethyl)-1-piperidine carboxylic acid 1-methylpropyl ester (Bayrepel) against *Aedes aegypti* (Linnaeus), *Anopheles stephensi* Liston mosquitoes and the sand fly, *Phlebotomus papatasi* which are vectors of human diseases. When five females of each species were simultaneously exposed to one half repellent-treated (4.8×10^{-2}μmol/cm^2) and one half untreated skin beneath the opening of a 3x4 cm cell, they bit exclusively on untreated skin. When treated and untreated skin was covered with cloth to preclude tactile contact with skin surfaces, the insects bit only through cloth covering untreated skin. The tests proved that each species was exquisitely capable of avoiding the compounds in their close-range feeding behavior and this was accomplished by olfactory chemical sensing. When cloth, untreated and repellent treated, was placed over the skin all species avoided the compounds and fed through the cloth only where repellent was absent. In the presence of any the three repellents within a cell, biting responses were reduced by about 50% relative to controls. Thus, besides inhibiting feeding on repellent-treated skin, the compounds also exhibited spatial-repellent activity.

The influence of burning on the decomposition and insect succession of pig carcasses

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Recent casework raised the question whether burning has an influence on the decomposition and insect succession of bodies. This was investigated by dousing three pig carcasses with different volumes of petrol and burning them to different degrees (Slight = 1.5L Medium =5L, Heavily = 10L). The insect succession onto these carcasses was compared to a control carcass. The control carcass decomposed at the fastest rate with the heavily burnt carcass at the slowest rate. Adult insects comprised the following: At the control carcass Diptera (60.8%), Coleoptera (29.9%) and Hymenoptera (8.8%). At the slightly burnt carcass Coleoptera (55.8%), Diptera (38.6%) and Hymenoptera (5.4%). The dominant orders at the medium burnt carcass were Coleoptera (38.5%), Diptera (33.3%) and Hymenoptera (28%). At the heavily burnt carcass Coleoptera (51.4%) and Diptera (28.6%) were dominant. Hymenoptera (18.7%) did not have a direct influence on decomposition, since they functioned as predators. *Chrysomya albiceps* was the dominant Calliphoridae species at the control and medium burnt carcasses and *Chrysomya marginalis* at the slightly and heavily burnt carcass. Representative samples of larvae collected during decomposition revealed that *C. albiceps* larvae preferred to feed underneath the carcasses, with *C. marginalis* larvae feeding mostly inside and at the top of the carcasses.

Human behaviors and environmental factors elevating the risk of exposure to *Ixodes pacificus* nymphs in a hardwood forest

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The nymph of the western black-legged tick, *Ixodes pacificus*, has been implicated as the primary vector of the Lyme disease spirochete, *Borrelia burgdorferi*, in northwestern California. In 2002 and 2003, six human behaviors and environmental factors were evaluated as potential risk factors for acquiring nymphal ticks in a dense woodland in this region. The prevalence of *B. burgdorferi* and the causative agents of human anaplasmosis (*Anaplasma phagocytophilum*) and human monocytic ehrlichiosis (*Ehrlichia chaffeensis*) also were determined in nymphs that had been collected from subjects or by dragging leaf litter. Time-adjusted tick-acquisition rates demonstrated that sitting on logs was the riskiest behavior followed, in descending rank, by gathering wood, sitting against trees, walking, stirring and sitting on leaf litter, and sitting on litter. The number of ticks acquired appeared to be unrelated to the type of footwear worn. Nymphal ticks were active throughout the daytime over a wide range of micrometeorologic conditions. Overall, about 3-4% of nymphs were infected with *A. phagocytophilum* and *B. burgdorferi*, regardless of the collection method, versus none with *E. chaffeensis*. Co-infection with *B. burgdorferi* and *A. phagocytophilum* was detected for the first time in this life stage of *I. pacificus*. The public health implications of these findings will be discussed briefly.

Genetically Modified Mosquitoes: A Real World View

G.C. Lanzaro

The concept of controlling mosquito-borne diseases via the development and implementation of genetically modified strains has been under serious consideration since the early 1990's. Although this idea has been met with great enthusiasm in some quarters there remains substantial skepticism in others. The general concept and progress toward the development of such a strategy will be presented. Numerous roadblocks to the development of a GM mosquito plan remain, including (i) lack of gene drivers, (ii) lack of robust effector molecules and mechanisms to manipulate their expression, (iii) poor efficiency of transgenesis, (iv), and (v) no consideration of logistical problems associated with implementation in the field. These will be presented and prospects for overcoming them discussed. Aspects of the ecology and population genetics of targeted species will be discussed as they relate to the successful release and establishment of GM mosquitoes in nature.

Reverse chemical ecology: chemical prospecting of mosquito attractants and repellents

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For a few decades, chemical ecologists have been utilizing insect antennae as the sensing element or biodetector, in electroantennogram (EAG) and gas chromatography-electroantennographic detection (GC-EAD) assays for the identification of minute amounts of pheromones and other semiochemicals. The "molecular" equivalent of these electrophysiology-based "short cut bioassays" is the screening of attractants and repellents based on their binding affinities to odorant-binding proteins (OBPs). OBPs assist hydrophobic semiochemicals to cross an aqueous barrier in the insect sensilla (sensillar lymph) and reach odorant receptors (ORs). Both OBPs and ORs contribute to the high specificity of the insect olfactory system by forming two layers of filters. Although binding per se does not necessarily imply a physiological function, test compounds that do not bind to OBPs can be eliminated from further behavioral studies as they will never "see" the receptors. Reverse chemical ecology is a protein-based approach that requires the full identification of OBPs, cDNA cloning, functional expression of OBPs, and binding assays. In this presentation, we will discuss the latest development of a NIH-funded project* aimed at the development of better attractants and repellents for *Culex* species, the principal vectors of West Nile Virus.M

* NIH-National Institute of Allergy and Infectious Diseases (1U01AI058267-01).

Developing methodologies for a fungal biopesticide against *Boophilus* microplus, the cattle tick

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The Queensland department of primary industries and fisheries has been investigating fungal biopesticides for livestock ectoparasites as part of a wider strategy to reduce the reliance upon chemicals in livestock industries. Fungal biopesticides have been available for the control of agricultural and horticultural pests for over thirty years and methodologies and accepted protocols for the development of these fungal biopesticides are now well established. Current work with the cattle tick has shown that livestock pests present additional challenges, some of which are due to the inability to conduct *in vitro* assays on obligate parasites, the cost of *in vivo* experiments using animals and the different environments the fungus must operate in when invading the parasitic and free-living phases. In addition, the development of formulation and application methods suitable for penetrating an animal's hair coat and delivering a lethal dose of spores to ticks present further challenges. Solutions to these problems have sometimes been obvious and straightforward but mostly have been arrived at slowly and painstakingly. We have concluded that the development of methodologies for fungal biopesticides of livestock ectoparasites is a multidisciplinary exercise requiring a thoughtful balance of appropriate *in vitro* bioassays with on-animal investigations.

Increased densities of vector mosquitoes in anthropogenic environments in the Kapiti region in New Zealand and potential ecological mechanisms as to why

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Landuse changes, including deforestation, agriculture and urbanisation, have coincided with increases in mosquito-borne diseases worldwide. Anthropogenic environmental changes can alter mosquito populations by modifying the characteristics of breeding habitats, but we still poorly understand the ecological mechanisms involved. Mosquito breeding habitats were sampled in native forest, pastureland and urbanland in the Kapiti region, New Zealand, and 17 environmental characteristics were measured of each. Higher immature mosquito densities were collected from mainly artificial containers in urbanland and pastureland compared to mainly ground pools in native forest. Nine environmental variables were significantly different between landuses but none of these were significantly related to mosquito density. Principal components analysis partitioned environmental data into seven ecological types. One ecological type was significantly related to both landuse and mosquito density, and positively associated with ammonia, bacteria and dissolved organic carbon (DOC), and negatively associated with nitrate and predatory damselfly larvae. The findings of this study support those from prior surveys in warmer climes of greater vector disease risk in anthropogenically-modified environments because of compromised ecosystem integrity. Unlike most previous field-based work, this study also suggests important mechanisms involved; pinpointing mainly bacteria, DOC, nitrate and damselfly larvae as acting in combination to regulate vector mosquito density.

The final word on the genetic, evolutionary and taxonomic relationship of *Pediculus humanus* and *P. capitis*

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Body lice are thought to have diverged from head lice when humans began to wear clothes. This recent evolutionary divergence and incomplete lineage sorting has complicated assessments of whether head lice and body lice are separate species. We used microsatellite markers to test lice from hosts that were infested with head and body lice simultaneously, to see if the head and body lice had interbred. Our results showed that head and body lice had not interbred on a host. Assignment tests revealed that lice that had migrated among hosts always moved from head to head, and body to body, but never between heads and bodies. We conclude that head and body lice are separate species. We speculate that head lice are descended from the lice of neanderthals and modern humans, and that body lice are descended from the lice of modern humans that left Africa about 100,000 ya.

The search for natural repellents from human beings against mosquitoes

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Blood sucking insects are differentially attracted to human beings. This is likely to be caused by differences in the semiochemical profiles of individual human hosts. Despite substantial research into insect-human interactions, the responsible semiochemicals have not yet been identified. An explanation for this difference is that unattractive individuals either lack attractant chemical compounds or may produce compounds that 'mask' the activity of attractants. A special air entrainment technique was developed and used to collect volatile chemicals from human volunteers, and the extracts analysed by gas chromatography (GC), coupled GC-electroantennography (GC-EAG) and GC-mass spectrometry (GC-MS). For the first time electrophysiological responses of female yellow fever mosquitoes, *Aedes aegypti* to volatiles from air entrainment extracts were recorded, and a number of electrophysiologically active compounds were identified. Previous behavioural studies have confirmed the differential attractiveness of volunteers to biting insects. A quantitative and qualitative analysis of compounds within the extracts, from individuals known to be differentially attractive to biting insects, has revealed significant differences in chemical profiles. The identification of behaviourally active compounds could lead to new and improved control technologies, whereby 'masking' compounds could potentially be incorporated into new, safe and natural repellents against biting insect pests.

Anopheles arabiensis at the edge of its distribution in northern state, Sudan

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Northern state, Sudan is mainly desert, rainfall is rare and most life is found close to the Nile. *Anopheles arabiensis* is the only anopheline and malaria vector found in the area in recent decades. It is present along the Nile downriver to about 200 km south of the Egyptian border, where poor terrain and control efforts prevent its spread further north. Initial results are presented from field and molecular studies to determine the population structure and dynamics of the species and related environmental factors. The receding Nile following the flood contributes most to sustaining *A. arabiensis*. Larval breeding sites can be found on low lying riverbank, ied-up inlets, or channels around islands, through most of the year. Land use is mainly associated with irrigation, but this does not appear to contribute greatly to anopheline numbers, however the extent of human presence and activities do increase the potential for anopheline breeding. Upriver from the fourth cataract and downriver from the third cataract, the terrain is rocky desert and less suitable for both humans and mosquitoes. The *An. arabiensis* population is thus semi-isolated within an approximately 700 km corridor a few kilometres wide and breaks down further into pockets depending on particular river features.

Control of biting midge (Ceratopogonidae) attack on residents of a marine canal estate in Redcliffe (Australia) by residual barrier treatment of Bistar 80SC® to ornamental garden plants in house yards

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Biting midge (Ceratopogonidae) breeding in marine sediments associated with engineered canals in a residential community in Redcliffe Australia have been responsible for considerable biting attack and nuisance. Limitations on approved applications of larvicides in breeding sites has historically restricted control to personal protection, midge screening and education of residents. In October 2003 – Feb 04, Redcliffe City Council funded a trial to test the efficacy of Bistar 80SC® (FMC Chemicals Pty Ltd) as a 0.1% AI (Bifenthrin) residual application to ornamental garden vegetation and dwelling external walls. Eight paired randomised treated and untreated premises were assessed for differences in biting midge exposure. The assessment was undertaken using CO_2 and Octenol baited light traps placed in treated and untreated yards. A self administered questionnaire scoring individual household daily experience of biting midge activity was also used. Untreated control premises received a placebo application of water alone to control bias in the resident questionnaire. Analysis of difference in light trap collections between treated and untreated showed a 76.4% reduction in midge (primarily *Culicoides molestus*) from treated premises. Resident survey results indicated occupants of treated premises experienced generally favourable biting midge conditions while residents from untreated homes experienced fluctuating exposure and discomfort corresponding to lunar emergence cycles.

Aerial efficacy assessment of Anvil 10+10 against *Anopheles quaimaculatus*

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Aerial ULV applications of Anvil 10+10 were conducted against caged, field-collected *Anopheles quaimaculatus* adults using Beecomist® 360A rotary atomizers. Standard and optimized applications were conducted at 0.62 fl oz (0.0036 lbs a.i.) per acre using stacked passes 3,500 ft and 5,000 ft upwind of the target site. Efficacy of Anvil 10+10 and impact of wind velocity at flight altitude (150 ft) were assessed by bioassay and analysis of collected droplets.

The potential role of aquatic insects in the transmission of buruli ulcer disease in Africa

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Buruli ulcer is the third most frequent mycobacterial disease in humans, next to tuberculosis and leprosy, often causing serious skin ulcerations, bone deformities, and disability. Unlike TB and leprosy, Buruli ulcer has emerged dramatically since the 1980's, with increasing numbers of cases being reported from at least 31 countries, mostly in tropical and subtropical areas, including Africa. Although poorly understood, the epidemiology of Buruli ulcer associates the resurgence with people who live and/or work close to wetlands, especially slow flowing (riverine) or stagnant water bodies (marshes, swamps), often created as a result of some human environmental disturbance. The disease is not known to be contagious between humans, and epidemiological studies have not yet established natural reservoirs or modes of transmission. However, it was recently demonstrated experimentally that *M. ulcerans* could survive and multiply exclusively within salivary glands of aquatic bugs (Naucoridae: *Naucoris* sp.) and they were able to transmit *M. ulcerans* to mice by their bites, suggesting a possible mechanism for human infection. The organism has been found in samples of water, biofilm and detritus. A possible trophic scenario for the natural mode of transmission of *M. ulcerans* is presented based on preliminary work in Ghana by the authors.

Efficacy of a Spinosad Suspension Concentrate Formulation for Controlling Chewing and Sucking Lice on Cattle Under Field Conditions

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Field studies were conducted with cattle in Wisconsin and Illinois between January and April, 2000, to determine the efficacy of 0.04% spray and 2 mg/kg pour-on treatments of a spinosad suspension concentrate formulation against chewing and sucking lice. Positive control substances included in the study were Co-Ral® EC (5.8% coumaphos) and CyLence™ Pour-on (1% cyfluthrin). Animals in the negative control group remained untreated. All lice species were visually sampled on each animal at weekly intervals after treatment, for eight weeks. Individual louse counts were summed over each examination site for each species. A repeated measures analysis using a mixed model (SAS PROC MIXED), was conducted for data pooled across both study sites. Both spinosad treatments and CyLence™ Pour-on provided excellent efficacy (98-100%) against *Bovicola bovis* over the entire study period; Co-Ral® EC efficacy opped below 90% from Weeks 6-8. The efficacy of the spinosad spray treatment against *Linognathus vituli* was >93% for the entire study and >88% for the spinosad pour-on treatment. Co-Ral® EC was >90% efficacious against *L. vituli* during Weeks 1-7, while CyLence™ Pour-on only exceeded 90% for Weeks 3-7. *Solenopotes capillatus* were found only at the Wisconsin site; all treatments were highly efficacious throughout the study period.

The effect of human age on dengue seroconversion risk in Iquitos, Peru

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Basic models of infectious disease often assume that force of infection is independent of host age. This assumption has been shown to be violated for many directly transmitted diseases, but has rarely been examined in vector-borne systems, such as dengue. Data from DNA fingerprinting studies of mosquito bloodmeals indicate that the biting rate of *Aedes aegypti* increases with human age. If risk for dengue infection increases with human age, models assuming an age-independent risk will underestimate the proportion of the population that needs to be vaccinated in order to eliminate disease transmission. A cohort of approximately 3,000 schoolchildren and their families in Iquitos, Peru were followed for up to 5 years (1999-2003) with blood samples collected at 6-month intervals. Sera were tested for dengue virus antibodies by the plaque reduction neutralization test. The effect of human age on risk for dengue seroconversion was evaluated using a multiple logistic regression model, which also included area of residence within the city. Preliminary analysis suggests that risk increased with age and was highest in the southern region of the city. Estimates of age-specific forces of infection can be used to design an optimal immunization program.

Oviposition Site Selection by Mosquitoes in Response to drought

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Mosquitoes are vectors of many important medical and veterinary diseases. Although outbreaks of mosquito borne disease can be correlated to changes in precipitation in the very short term, precipitation rarely corresponds to mosquito abundance in the long term. Here, I present the results of a field based mesocosm experiment designed to test how water level and drought influence the oviposition behaviour of two species of mosquitoes. I used two water levels (high and low) and two drought treatments (normal and drought). Water for the drought treatments was obtained from ponds that had experienced ying for a period of a month prior to experimentation. The two mosquito species differed markedly in their oviposition response. The egg laying behavior of *Ochlerlatatus notoscriptus* was significantly greater in the drought treatments regardless of water level. However, the egg laying behavior *Anopheles annulipes* showed the opposite response with significantly more larvae present in the high water treatments regardless of whether the treatment water was normal or in drought. My results provide supportive evidence that mosquitoes may show population outbreaks following drought years and that this framework may provide a reliable way of predicting year-to–year variation in mosquito abundances based on individual species oviposition response.

Aedes aegypti **surveillance in Iquitos, Peru: a 4- year test of the pupal/demographic survey**

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We present results from 4 years of pupal/demographic surveys of a circuit of ≈ 6,000 houses surveyed >10 times in the Amazonian city of Iquitos, Peru. We compare traditional larval indices with pupal and adult mosquito indices. We evaluated *Aedes aegypti* production from different container classes, including those based on water management practices overtime and space. A total of 289,941 water-holding containers were characterized, of which 7.3% were positive for *Ae. aegypti*. Although, temporal and geographic variations were detected for all variables examined, consistent production patterns were observed for the lid status (lids: 32% wet containers, 2% pupal production), container location (outdoor: 43% wet containers; 85% pupal production) and method by which the container was filled with water (rain filled: 15% wet containers; 88.3% pupal production). Based on these patterns and additional targeting of containers with highly variable pupal counts we propose a targeted control strategy for Iquitos that would reduce *Ae. aegypti* populations below published but untested threshold values for Iquitos.

Screening medicinal plant extracts for effects on *Aedes aegypti* (Diptera : Culicidae) and toxicity to a non target organism

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A preliminary study was conducted to find out the effects of the extracts of 112 medicinal plant species, collected from the southern part of Thailand, on *Aedes aegypti*. Studies on larvicidal properties of plant extracts against the third and fourth instar larvae of *Ae. aegypti* showed fourteen species with high toxicity. Six plant species were comparatively more effective against the larvae at very low concentration. These extracts demonstrated no or very low toxicity to guppy fish (*Poecilia reticulata*). Three plants with promising larvicidal activity, *Mammea siamensis*, *Anethum graveolens* and *Annona muricata,* were used to study of the impact of the extracts on the life cycle of *Ae. aegypti*. The results showed that these plants affected the reproductive potential of surviving adults by reducing the number of eggs laid and egg hatchability. The first instar larvae were very susceptible to *A. muricata* and the second instar larvae were susceptible to *A. graveolens* while the third and fourth instar larvae were susceptible to *M. siamensis*. These extracts delayed larval development and inhibited adult emergence.

Tick vector-pathogen interrelationships

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The interrelationships between tick-borne pathogens, their tick vectors and diverse vertebrate hosts, are represented by a triangle of parasitic interactions. The interactions are between (i) tick-pathogen, (ii) pathogen-host, and (iii) tick-host. In (i) the pathogen interacts with its vector, infecting and replicating within tick cells or extracellular spaces. The ability of a particular tick species to act as a vector depends on whether the pathogen can survive and overcome several 'barriers' within the tick. However, this is a simplistic view of the tick vector-pathogen interrelationship as both (ii) and (iii) influence the success of (i). This triangle of interactions is focused at the skin site of tick feeding - the initial site at which pathogens gain access to either their host or vector. For pathogens, the skin-feeding site is an ecologically privileged niche, profoundly modified by the pharmacological activities of bioactive tick saliva molecules. Exploitation of this niche is known as saliva-activated transmission (SAT). Tick saliva molecules that promote SAT have not been identified but appear to differ for different tick vector-pathogen interrelationships. Identifying the SAT factors and their mode of action offers new insights for controlling ticks and tick-borne diseases.

Argentina: veterinary forensic entomology as a potential tool against rustling

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Cattle rustling entails economic loss to cattle owners and danger to their safety, marketing of uninspected meat and hides and loss to the State from unpaid taxes. In 2003, the Faculty of Veterinary Medicine (Tandil, Buenos Aires province, Argentina) incorporated forensic entomology to a course of legal veterinary medicine aimed at fighting rustling. Preliminary field experiments showed a large proportion of hemisynanthropic insects, most of them Neotropical, which may represent the autochthonous necrophagous fauna, now displaced by synanthropic introduced species in urban environments. Colonization and development times proved longer in the arid climate of Tandil than at Buenos Aires city, where most published data were gathered. Field experiments whith bovine bodies and parts are required. Hitherto, bovines have been avoided as a model because of the added complication of the ruminant digestive tract. Since most countries with rustling problems also have a pattern of introduced species displacing the original necrophagous fauna in city environments, they might benefit from knowledge obtained through these studies.

Arboviruses, mosquitoes, and RNA interference

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The identification of critical arbovirus-vector interactions reveals new insights into vector competence. For instance, flavivirus and alphavirus replication in mosquitoes triggers a natural RNA interference (RNAi) response; however, viral counterdefenses probably allow RNAi evasion and establishment of persistent infections. We can enhance RNAi to dengue virus type 2 (DENV2) in *Aedes* species by administration of double-stranded (ds) RNA derived from the DENV2 genome, either by direct transfection of dsRNA, transient expression from a Sindbis virus (SINV) transducing system, or stable transformation with a plasmid that expresses a hairpin-forming transcript. RNAi is sequence-specific, is maintained by low concentrations of effector RNA, blocks accumulation of intracellular viral RNA, and results in the appearance of 21-25 nt siRNA homologous to the mRNA target. We have identified genes from the Dicer and Argonaute families in 2 mosquito genera and have used RNAi to knock down their expression to determine which genes have a role in virus resistance. We have evidence that DENV2 and some strains of SINV establish persistent infections in mosquitoes by suppression of RNAi, and are investigating mechanisms. We are identifying viral-encoded suppressors by site-specific mutations in infectious cDNA clones. Our work suggests that persistent infections in mosquitoes result from a balance between host RNA silencing and viral counterdefenses and provides new strategies for tipping the balance in favor of the vector host.

Endosymbionts and sex ratios in the human head louse

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At the turn of the 19th century the first observations of a female-biased sex ratio in broods and populations of head lice had been reported. A study by Buxton in 1938 on the sex ratio of lice on prisoners in Ceylon is still today the subject of reanalyses. However, in the last sixty years no data have been collected, especially on more normal levels of infestations. Here we report a female bias of head lice in a survey of 480 school children in Argentina. This bias is independent of lice intensity. These lice as well as lice from pupils in Britain were carrying several, related strains of the endosymbiotic bacterium *Wolbachia*, one of the increasing number of intracellular sex ratio distorters. No lice without *Wolbachia* were detected. Similar *Wolbachia* strains are also present in the pig louse suggesting that this endosymbiont might have marked influence on the biology of the whole order. The sex ratio distortion in head lice that has been reported world wide is stable over time; that is a remarkable deviation from the stability of frequency-dependent selection of Fisher's sex ratio.

Blood Feeding in *Cnephia* (Diptera: Simuliidae): Molecular Phylogeny

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Five species of *Cnephia* were studied, three non-biting, one a mammal feeder, and one a bird feeder. Of the non-feeding species, *C. eremite* is Holarctic in distribution and specimens were examined from both Canada and Sweden. *Cnephia pallipes* is a Palearctic non-feeder and *C. dacotensis* a temperate Nearctic non-feeder. *Cnephia pecuarum* is an economically important large mammal feeder in south-central U.S. and *C. ornithophilia* a widely distributed bird feeder in southern and eastern North America. Sequence was obtained from the complete mitochondrial CO-II gene (687 bp) and partial EF1-alpha nuclear gene (1030 bp). Molecular data is discussed in relation to conventional wisdom about relationships among species based on cytological and biological information, geographic distribution and genetic diversity within species. Molecular data strongly support *Cnephia* as a monophyletic clade in the tribe Simuliini. Speciation hypotheses were not fully concordant between molecular and conventional data.

Onchocerciasis in Central America: Ecology & Control

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Detection of O. volvulus larvae in vector populations of black flies is of prime importance in the assessment of the effectiveness of ivermectin control programs. Detection of larvae is attained by dissection of individual flies or by PCR (amplification of an Onchocerca 150 bp repeated sequence, followed by hybridization of the PCR products with a species-specific DNA probe) from pools of flies. The PCR was compared with dissection method to estimate the infection rate (p) and the mean larval load (any larval stage) per parous fly (m) in a local vector population. Both methods produced statistically similar estimates, thus PCR is an alternative and efficient method for entomological surveillance. Sample sizes required to estimate m from p for fixed levels of precision were calculated using Mexican and Guatemalan data, assuming that k (an aggregation parameter from the larval distribution among parous flies) is known. Optimal sampling and PCR was used to evaluate the impact of ivermectin on m, p and transmission potentials in Mexico. The data suggest that substantial progress towards the goal of suppressing transmission has been accomplished. In southern Chiapas, entomological estimates decreased in comparison with the baseline data, but PCR positivity was still detectable. In northern Chiapas and Oaxaca, with no baseline data, there was an absence or near absence of infected and infective flies.

New and resurgent arboviral disease threatening humans in Australia

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Australia has a range of indigenous arboviruses and an introduced few that are of public health concern; all are mosquito-borne. Endemic encephalitogenic viruses (Murray Valley Encephalitis, Kunjin) are annually active and responsible for occasional cases in the northwest but, despite widespread vector distribution, only rare activity and no epidemics in the southeast since 1974. The most common arboviruses, Ross River annually infecting many thousands nation-wide causing polyarthritis, and Barmah Forest (a similar syndrome) on the increase and responsible for outbreaks of hundreds regionally, have a spectrum of coastal saline and inland freshwater vectors. The most important exotics are the returning dengue viruses with outbreaks of hundreds confined to Queensland by vector distribution but with virus endemicity and vector spread threatening, and the recently introduced Japanese Encephalitis virus causing only a few deaths in northern Queensland but with virus persistence and its affinity for a local vector threatening. Environmental factors drive arbovirus activity by influencing vector populations, which vary with virus and circumstance, although little is known of the role of reservoir hosts. Tourism and development activities that increase human/vector/virus contact, and climate changes that promote vector and virus proliferation, threaten to exacerbate the disease threat to the public health.

Repellents for integrated tsetse management

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ICIPE is in the forefront in identifying potent repellents, both synthetic analogues of natural tsetse repellents and natural repellent blends from un-preferred animals like waterbuck and zebras. One potent synthetic repellent for savannah tsetse species, developed by molecular optimization studies, acts as an olfactory antagonist of a key phenolic kairomone, which flies use to locate the host for feeding (Saini & Hassanali; patent application). This repellent substantially reduce tsetse challenge (> 80%) and feeding efficiency (>80%) on cattle. A prototype dispenser which is placed around the neck of an animal has been developed. Field trials indicate that the repellent can provide effective protection to cattle but all animals in a herd need to be protected or else significantly higher infection occurs in unprotected animals. Experiments are ongoing to evaluate how repellents can be integrated with other existing tsetse control techniques. A 'push-pull' strategy that deploys repellents (to 'push') and baited traps / targets (to 'pull') to enhance suppression rates is being evaluated. A variant of this may involve the use of a proportion of cattle with 'pour on' insecticides as live baits. A business plan for commercial production and distribution of repellent products is also being developed.

Mitochondrial genome organization of lice is highly variable

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Mitochondrial (mt) genome organization is extremely conserved in mammals, vertebrates and most arthropods, e.g., humans and frogs have the same mt genome organization; so have fruit flies and water fleas. We have sequenced the entire mt genomes of the wallaby louse (Phthiraptera: Amblycera) and the pigeon louse (Ischnocera), and part of the mt genomes of the human louse (Anoplura) and four other amblyceran lice. Unlike most arthropods, the mt genome organization of these lice is highly variable. Lice share only 1-2 gene boundaries with fruit flies. No gene boundaries were shared among lice of the three suborders. Mt genome organization differs even among three genera within the suborder Amblycera. Changes in the mt genome organization of lice include gene translocation, inversion, separation of rRNA genes and loss or multiplication of non-coding regions. In addition to lice, we found mt genome organization was variable or highly variable in other two hemipteroid orders, Psocoptera (psocids) and Thysanoptera (thrips), but is strictly conserved in the order Hemiptera (bugs). We speculate that the changes in the mt genome organization in lice, psocids and thrips may be related to their life-history traits such as small body size, small colony size, and high degree of inbreeding.

West Nile virus activity in the USA and regional differences in vectors and transmission dynamics

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In the USA, West Nile virus (WNV) was first detected in New York City in 1999. By the fall of 2003, WNV had spread south and west to 45 states. During 1999–2003, 13,405 human cases of WNV disease were reported. Principal enzootic and epidemic vectors of WNV in the USA are *Culex (Culex)* mosquitoes. In the Northeast and upper Midwest, *Cx. pipiens* L. including form molestus, *Cx. salinarius* Coquillett and *Cx. restuans* Theobald are the most important vectors. In the South, *Cx. quinquefasciatus* Say and *Cx. salinarius* are the most important vectors. In Florida and the southern portions of LA, MS, and AL, *Cx. nigripalpus* Theobald is a potential vector. In the Rocky Mountain and High Plans states, *Cx. tarsalis* Coquillett and *Cx. pipiens* are the primary vectors. In the arid Southwest, *Cx. tarsalis* is the primary vector with *Cx. quinquefasciatus* playing a role in urban areas. Across much of the mid-latitudes, *Cx. pipiens* and *Cx. quinquefasciatus* hybridize and both taxa and hybrids are important vectors. California will experience WNV disease in the future and important vectors are likely to be *Cx. tarsalis* and members of the *Cx. pipiens* complex, with *Cx. stigmatosoma* Dyar and *Cx. erythrothorax* Dyar being focally important. Important avian host vary with regional abundance, however, the house sparrow (*Passer domesticus*) appears important in virus transmission throughout the USA.

Measures of *Aedes aegypti* density and the risk of dengue virus transmission.

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In two large, cohort-based prospective studies we are defining entomological risk and characterizing its relationship to human dengue infection. Based on results from more than 5 years of field research in the Amazonian city of Iquitos, Peru we are determining the appropriate scale (spatial and temporal) at which to measure entomological risk and dengue virus transmission. A new research program in Khamphaeng Phet, Thailand is an extension of the Iquitos project and will explicitly link in time and space various measures of entomological risk to human dengue infections

Aedes aegypti **genomics**

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"We have initiated a genome project for *Aedes aegypti* that will ultimately lead to complete sequence information. We are sequencing both ends of genomic DNA Bacterial Artificial Chromosome (BAC) clones and random cDNAs representing a variety of tissues, and physically mapping selected BAC clones. BAC clone end-sequencing is completed and the data are available at GenBank. We plan to sequence and analyze both ends of ~80,000 cDNAs derived from normalized libraries produced from 4 specific mRNA sources and one library enriched for full-length sequences. Individual libraries are in various stages of construction, with one competed (*Brugia malayi*-infected) and 29,824 ESTs generated, and sequencing of a second (*Plasmodium gallinaceum*-infected) nearly completed. The third library (females exposed to a dengue virus) has just been completed and we are in the process of constructing libraries from 1) fat body and 2) combined developmental stages. We have incorporated all sequences from our genome project with gene sequences available in GenBank into an *A. aegypti* Gene Index available at TIGR (www.tigr.org/tdb/tgi/aegi/). We have recently (2/13/2004) been notified that NIH-NIAID has selected *A. aegypti* for full genome sequencing. We anticipate this will include an 8X coverage that should provide a high quality genome assembly.
"

Mitochondrial genome organization of lice is highly variable

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Mitochondrial (mt) genome organization is extremely conserved in mammals, vertebrates and most arthropods, e.g., humans and frogs have the same mt genome organization; so have fruit flies and water fleas. We have sequenced the entire mt genomes of the wallaby louse (Phthiraptera: Amblycera) and the pigeon louse (Ischnocera), and part of the mt genomes of the human louse (Anoplura) and four other amblyceran lice. Unlike most arthropods, the mt genome organization of these lice is highly variable. Lice share only 1-2 gene boundaries with fruit flies. No gene boundaries were shared among lice of the three suborders. Mt genome organization differs even among three genera within the suborder Amblycera. Changes in the mt genome organization of lice include gene translocation, inversion, separation of rRNA genes and loss or multiplication of non-coding regions. In addition to lice, we found mt genome organization was variable or highly variable in other two hemipteroid orders, Psocoptera (psocids) and Thysanoptera (thrips), but is strictly conserved in the order Hemiptera (bugs). We speculate that the changes in the mt genome organization in lice, psocids and thrips may be related to their life-history traits such as small body size, small colony size, and high degree of inbreeding.

Simuliid taxonomy in relation to transmission of human onchocerciasis and mansonelliasis in Brazil.

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Mansonelliasis was recorded as widespread in the southern Amazon over 50 years ago and human onchocerciasis 35 years ago in a discrete area in the northern Amazon region of Brazil. Incrimination of the simuliid vectors that transmit the filarial parasites responsible for these diseases has only been possible during the last three decades, largely due to the logistical difficulty in collecting species in these regions and the complex taxonomy of members of the *Simulium amazonicum* group to which several belong. This has been caused by the morphological similarity between vector and non vector species of the group. The use of digital image analysis has facilitated the morphological definition of species in the group, recently augmented by cytological and molecular studies. Our current classification of the group based on these techniques is presented together with distribution maps of vector species that suggest that disease dispersal to non endemic areas may be occurring as infected individuals migrate from the disease foci as rural areas of the Amazon are developed.

Polymerase chain reaction-based differentiation of members of the *Anopheles culicifacies* complex

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Anopheles culicifacies, the principal vector of malaria in India, is responsible for over 65% of total malaria in India and is a complex of 5 cryptic species designated as species A, B, C, D, and E. These members are morphologically indistinguishable at any stage of life and can be identified by examining species-specific inversions in polytene chromosome and mitotic karyotype of Y-chromosome of male progeny. In an attempt to develop a simple alternative to cyto-taxonomy, we have developed an Allele-Specific Polymerase Chain Reaction (ASPCR) assay targeted to D3 domain of 28S ribosomal DNA, which unambiguously differentiates *An. culicifacies* species A and D from species B, C, and E. The assay was evaluated using chromosomally-identified specimens of *An. culicifacies* originating from different geographical regions of India representing different sympatric associations and was found to be specific. The culicifacies-specific primers used in the assay did not cross-react to other anophelines as tested against all members of *An. fluviatilis* complex (species S, T, and U), *An. minimus* (species A), *An. stephensi* and *An. subpictus.* The search of genomic database using culicifacies-specific primers sequences revealed that these primers are specific to *An. culicifacies* only. The possible use of this diagnostic assay in disease vector control programme for discrimination of vector species from non-vector will be discussed.

Insecticides don't kill lice but formulations of insecticides do!

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Head lice treatment preparations are labelled according to the qualitative and quantitative content of their active ingredients. A recent review of this group of products by the Medicines Evaluation Committee of Australian Therapeutic Goods Administration concluded that "There is a paucity of good quality information on the effects of different formulations on the efficacy of head lice products". This presentation will review the available evidence and identify some of the issues which may impact on dosage form design of head lice preparations.

Potency and avidity of tick paralysis antitoxin in Australia

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Tick paralysis due to the Australian paralysis tick, *Ixodes holocyclus* is a potentially-lethal syndrome affecting domestic pets, livestock and humans along the east coast of Australia mainly during late spring and early summer. There is no vaccine available at present. Prevention in animals largely depends on acaricidal treatment. Treatment of paralysis is based on a commercially-available antiserum, raised in hyperimmune dogs by progressive and cumulative antibody stimulation by repeated infestation with adult female ticks. This serum constitutes a specific, antitoxin widely used by veterinarians and to a limited extent by physicians to treat cases of tick paralysis. Standardisation at the minimum arbitrary level of 500 antitoxin units per ml is legally required. Variability occurs in potency and particularly in avidity (the well-known relative variability on dilution to neutralise an active toxin [in this case, holocyclotoxin] *in vivo* and *in vitro*). A modified method using dilution of antitoxin as opposed to the earlier method of dilution of toxin, may be used to study this variability. This variability will be discussed and possible explanations proposed.

Global expression profiling of the major detoxification enzyme families in the major African malaria vector *Anopheles gambiae*

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Vector control programs based on the use of insecticides, whether it be by residual house spraying or treated bed nets, are being challenged by the emergence of insecticide resistance. For example, there is wide scale resistance to pyrethroids in mosquitoes from West Africa, which are the major insecticides used in malaria control. One reason for this is the increased detoxification of insecticides by three major enzyme families; glutathione *S*-transferases, cytochrome P450 monooxygenases and the carboxylesterases. The recent publication of the complete genome sequence of the major malaria vector *Anopheles gambiae,* has allowed us to construct a highly specific 'detoxification chip', which contains fragments of all the genes putatively implicated in insecticide resistance in this mosquito. We have used microarray technology to monitor the expression of genes in insecticide-susceptible and resistant mosquito strains to pin-point the genes responsible for resistance. Furthermore, to generate a more detailed profile, their expression has also been assessed in the various life stages of the mosquito and in male and female insects. The detoxification chip is an excellent tool to provide a platform with which to go on and determine the function of these enzymes.

Odour blends in the host-seeking behaviour of the malaria vector *Anopheles gambiae*.

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The mosquito *Anopheles gambiae* is the most important malaria vector in Africa primarily because of its strong anthropophilic behaviour. We have studied factors affecting host-seeking behaviour of this mosquito, notably the olfactory cues with which human hosts are being detected. Responses of the mosquitoes to human skin emanations were studied with electrophysiological and behavioural essays. We demonstrated that important olfactory stimuli for *An. gambiae* are ammonia, L-lactic acid and several aliphatic carboxylic acids. EAG and single sensillum recordings demonstrated significant dose-responses in olfactory cells of grooved peg and trichoid sensilla. Behavioural responses were observed with single and mixed odours in an olfactometer. *An. gambiae* always responded to ammonium, although as a single component ammonium could not compete with the natural odours from human skin. Ammonia was an essential synergist to cause activity of lactic acid and/or carboxylic acids. Laboratory results were complemented by field studies in West Africa, where it was found that wild *An. gambiae* were attracted to a synthetic blend of volatiles from a distance. The same blend of odours that caused high attraction in the olfactometer, attracted most mosquitoes to odour-baited traps. The results are discussed with consideration for the development of olfaction-based technologies for malaria control.

Novel mosquito repellents derived from essential oils of plants in Thailand

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Mosquito-borne diseases, such as dengue, malaria and filariasis are serious public health problems in many countries, especially in the tropical regions of the world. These diseases are caused by pathogens that are transmitted to man through mosquito biting only, thus personal protection from mosquito bites is then considered as one of the strategies to prevent them. Efforts to develop alternatives to replace chemical repellents have been increasingly considered, including extracting and evaluating plant products for repellent activity. The development and use of locally available plant products showing repellent activity thus avails an alternative strategy for prevention of mosquito-borne diseases. In this study we evaluated and reported repellent effects of essential oils extracted from plants against 4 mosquito vectors: *Aedes aegypti*, *Ae. albopictus*, *Anopheles dirus* and *Culex quinquefasciatus* under laboratory conditions using human volunteers. The essential oils were extracted from various parts of 22 plant species, belonging to 10 families, and the oils were prepared as 10% solution in absolute ethanol and some additives. The essential oils showing high degree of repellency were subsequently formulated as topical repellents and tested against day- and night-biting mosquitoes under field conditions. The results obtained from both laboratory and field evaluations will be presented.

Evaluation and development of cockroach repellents derived from essential oils of plants

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Cockroaches are common insects found throughout the world, especially in the tropical regions. They are generally recognized as not only household pests but also pose a high health hazard to humans. Various aspects of cockroach control including chemical and non-chemical methods have been researched on an ongoing basis. Repellents against cockroaches are considered as one of alternatives to eliminate cockroaches from dwellings and other harborage places. Attempts have been made to find and develop cockroach repellents derived from plant extracts. We evaluated 20 essential oils extracted from various plants such as basil, black pepper, eucalyptus, galanga, guava, kaffir lime, etc., for repellency against two cockroach species: the American cockroach (*Periplaneta americana*) and the German cockroach (*Blatella germanica*). The experiments were carried out against adult cockroaches in the laboratory. It was found that the tested essential oils provided various degrees of repellency against cockroaches, ranging from 40 to 100%. The essential oil extracted from kaffir lime leaves showed excellent activity, manifesting 100% repellency against both cockroach species. The essential oils providing high degrees of repellency (>80%) were subsequently formulated as cockroach repellents and tested against cockroaches in the field. Data from both laboratory and field tests will be presented.

Recent trends in louse control in the USA

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Since the advent of permethrin resistance among head lice (*Pediculus h. capitis*) in the 1990's in the USA, the hegemony of permethrin and synergized-pyrethrin products has given way to a more diverse array of louse control products. Many of the new products use 'natural' ingredients since consumers are anxious to avoid applying insecticides to their chilen, but efficacy is often questionable. Products for nit removal, for repelling lice and for louse detection are all available. The synergized-pyrethrins and permethrin-based products still have a dominant position. New active ingredients and novel technologies have been, and are, being evaluated for louse control – information on most are not yet publicly available. Products now on the market are discussed. FDA requirements are summarized Market size and product market-share are estimated. Future trends are predicted.

The *Simulium ochraceum* (species complex) (Diptera: Simuliidae) and its distribution in relation to onchocerciasis transmission in Latin America.

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Simulium ochraceum is the primary vector of the disease in Central America and it could be responsible for sporadic transmission of the disease in mainland Ecuador. It consists of three cytotypes which probably have different biting behaviour and susceptibility to the parasite. The morphological variation of this species along its distribution range and its rôle in onchocerciasis transmission are here discussed. The female of *S. ochraceum s.l.* is recognised by thorax orange with 1+1 white pruinose, sub-median cunae (light source anterior). Within the complex, variation in the length of white pruinose vittae and coloration of postnotum and legs occur. *Simulium ochraceum s.l.* shows marked differences in its biting behaviour, being anthropophilic in Central and South America and largely zoophilic in the Caribbean Islands. It bites man occasionally in Brazil and Venezuela. The use of integrated taxonomic techniques is suggested to discover the meaning of morphological variations in terms of cytotypes. The host capacity of *S. ochraceum s.l.* is low because of the presence of cibarial teeth, but it is an efficient vector in hyperendemic areas for the disease. Other factors affecting the vector capacity of this species in the transmission of onchocerciasis in Latin America are discussed.

Emergence of epidemic dengue in Kerala State, southern India, with an account of establishment and dominance of the vector mosquito *Aedes albopictus* (Skuse)

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Dengue (including dengue hemorrhagic fever and dengue shock syndrome) has emerged as a major health hazard in India in recent decades as it is being increasingly reported in epidemic proportions from even States hitherto largely *terra incognito* for epidemic dengue, e.g. Kerala State in south India. Several focal outbreaks of dengue fever have been reported from Kerala in recent years, originating in the sylvatic and mountainous terrains of Kottayam, Idukki and Ernakulam, and spreading to coastal districts. The enormity of problem is gauged from the fact that the incidence hiked steeply from merely 14 cases and 4 deaths in 1997 to 3377(241-fold increase) and 66 (>16-fold increase) by November, 2003. While the principal dengue vector, *Aedes aegypti*, was insignificantly prevalent in density mostly along the coastal belt urban areas, *Ae. albopictus* has dominated in prevalence (71.2%), followed by *Armigeres subalbatus* (20.7%), *Aedes aegypti* (4.4%) and *Culex uniformis* (1.5%) amongst all the 22 species under seven genera occurring in the State. Twice the dengue virus had been detected in *Ae. albopictus* adults reared from the field-collected immatures from Western Ghat region. *Aedes aegypti* is, however, yet to be incriminated from Kerala State.

The truth is out there: application of new technology for the understanding and surveillance of mosquito-borne arboviruses

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Since 1995, Japanese encephalitis virus (JEV) has emerged as an annual threat to northern Australia. To monitor for future incursions of JEV, a sentinel pig programme was established in the Torres Strait and on Cape York Peninsula. While this system has been successful in detecting the virus, it is not ideal: deployment of the pigs is expensive, dangerous to handlers, and, as pigs are the primary amplifying hosts of JEV, they could serve as a source of virus. We outline the development of a mosquito-based sentinel system for the detection of JEV activity in remote areas of northern Australia. This approach is based on application of new generation mosquito traps, such as the propane-powered Mosquito Magnet™, for collection of mosquitoes, and a real time TaqMan RT-PCR for specific detection of viral RNA. In a field trial of the system, it was demonstrated that a single JEV-infected mosquito could be detected in pools of <5000 mosquitoes, maintained for <14 days in tropical conditions. Since 2002, 10 mosquito pools have been positive for JEV at the same time sentinel pigs have seroconverted to the virus. The relative efficacy of the mosquito-based system for detection of JEV and other arboviruses will be discussed.

The New Year's party that went terribly wrong – A case study on the influence of wrapping on decomposition of a body.

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In this study, an example of unusual disposal of remains is presented with comments on the changes in the resulting patterns of arthropod invasion. A young female disappeared the night of 31st December 2001, after a New Year's party. The evening of 12th January 2002 a suspect was arrested and the body was found. The body was in an advanced stage of decomposition, almost skeletonized. The insects present were late third instar *Chrysomya albiceps* larvae, a few adult *Dermestes maculates* and a few adult Piophilidae. These insects did not correspond to the stage of decomposition. Diptera pupae were also expected. The question arose whether the body might have been kept for a few days, excluding insects, before disposal. It was later established that the body was wrapped in a cotton sheet before it was left in the field. The crime scene was reconstructed, using two pig carcasses, the one wrapped in a sheet and the other unwrapped. Decomposition and insect succession of the two carcasses were monitored and compared. The insects present on the carcasses were the same, but the wrapping of the carcass slowed down the decomposition process, although colonization of the two carcasses occurred at the same time.

The colonization of buried remains at different depths

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A body exposed in the environment represents a trophic resource for necrophagous organisms. Faunal succession and free access to the corpse are influenced by environmental factors and by the state of the body remains. Instead, in the case of buried corpses, few organisms are able to reach and process these remains. To study which insect species are responsible for underground degradation, we performed experiments by burying some boxes with meal at different soil depths. Two experimental areas in NE Italy were chosen: one in a hilly landscape without agricultural activities, the other in a zone with high human exploitation. In these sites both grassland and deciduous forest were tested. The buried boxes were collected at different times and insect specimens were identified. Diptera belonging to 8 families were found: Stratiomyidae, Phoridae, Sphaeroceridae, Piophilidae, Anthomyiidae, Muscidae, Sarcophagidae and Calliphoridae. The most abundant being the Phoridae, with *Diplonevra funebris* the most abundant species (80% of the total). Calliphoridae and Sarcophagidae were present only in the non-buried boxes. Several Coleoptera were also collected: Staphylinidae were found at all the depths, both as adults and larvae, while Silphidae and Dermestidae were collected only in the non-buried boxes.

Bionomic of *Anopheles stephensi* Liston in a malarious area, Hormozgan province, southern Iran, 2002

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A total of 25000 malaria cases has been reported from Iran. From which Hormozgan province has a higher API comprising 30% of total cases. *Anopheles stephensi* is an important malaria vector in Iran. This species has two activity peaks, one in spring and another in autumn in the region. Mosquitoes of this species prefer to rest and feed indoors. Larval habitats are quite different. All three known biological forms of this species; type, intermediate, and *Mysorensis* occur in the province. Insecticides susceptibility tests showed susceptibility to bendiocarb, propoxur, malathion, fenitrothion, deltamethrin, permethrin, cyfluthrin and lambdacyhalothrin, but resistant to DDT and low level of tolerance to dielin. Larvicidal activity of malathion, fenitrothion, temephos and chlorpyrifos at diagnostic dose exhibited susceptibility to all larvicides except to fenitrothion. The irritability tests to pyrethroid insecticides showed a high level of irritancy to permethrin and lambda-cyhalothrin, but low irritancy to cyfluthrin and deltamethrin.

Identification of Australian carrion-breeding blowflies using multiple gene regions

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"Carrion-breeding blowflies have well recognised forensic importance. Their period of development on a corpse can help indicate the time elapsed since death. The applied importance of these flies makes their reliable identification a necessity. Mitochondrial DNA markers may help identify immature blowflies whose identity is difficult or impossible to confirm morphologically. DNA recognition of Australian blowflies poses a special challenge, because their relatively recent speciation makes it necessary to focus on highly variable gene regions for their molecular separation. Thirty-four blowfly species from southern and north-eastern Australia were included in our study, representing almost all species currently known or suspected to be carrion-breeders. Phylogenetic analyses of DNA sequences from the mitochondrial genes COI, COII and ND4 were conducted using parsimony, maximum likelihood and Bayesian methods. In general, these genes provide a powerful tool for the identification of the immature stages of Australian carrion-breeding blowflies. However, in some cases mtDNA sequence divergence is very low, even though statistical support for the separation of individual specimens may be high. Since this leaves open the potential for misidentifications, we have also investigated the applicability of variable nuclear gene markers, the ITS1 and ITS2 regions, to the molecular identification of very closely related species."

Emerging Vectors in Urban Wetland Ecosystems

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The use of constructed treatment wetlands for water quality improvement, water reclamation, wetland habitat supplementation, and other goals is increasing worldwide. Man-made wetlands can provide a cost-effective alternative to conventional water treatment facilities for municipal wastewater and are one of several technologies being used to improve the water quality of urban/suburban storm water runoff. A drawback of man-made wetlands used to treat municipal and storm water in areas of rapid human development is the juxtaposition of developmental sites for pathogen-transmitting and pestiferous mosquitoes with human development and peridomestic reservoirs of the causative agents of disease. Management practices and design features thought to be important for water quality improvement often enhance mosquito production. Vector control activities can be impeded by the management practices used to fulfill simultaneously the diverse goals for multipurpose constructed wetlands. The production of native mosquito species which are known to be vectors of the causative agents of disease remains a concern at man-made wetlands and at many best management practices for treating non-point pollutants in the urban environment. The introduction of novel pathogens and vectors into urban landscapes containing treatment wetlands creates new challenges for agencies charged with protecting public health.

The response of mosquito populations to conservation, rehabilitation and construction of urban wetlands at Sydney Olympic Park, NSW, Australia.

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Homebush Bay once contained diverse, yet degraded, estuarine and freshwater wetlands that were either removed or rehabilitated in conjunction with the Sydney Olympic Park urban redevelopment. In addition, extensive freshwater wetlands were constructed for water storage, waste-water treatment, wildlife conservation and aesthetic enhancement. Over 50 mosquito species belonging to 8 genera have been identified from the area, and the potential increases in nuisance biting and public health risks following redevelopment have been a major concern for local authorities. Poor tidal flushing of the estuarine wetlands resulted in unusually large populations of the saltmarsh mosquito, *Ochlerotatus vigilax*, a known virus vector and biting pest, but populations have declined following the commencement of pesticide application and wetland remediation programs. *Culex annulirostris* and *Coquillettidia linealis* are two freshwater pest/vector species that were abundant but declined with the redevelopment. Although the constructed freshwater wetlands were designed to minimise mosquito populations, they have become more productive with increasing vegetation density. Additionally, domestic species *Ochlerotatus notoscriptus* and *Culex quinquefasciatus* are expected to increase with the further residential development. This presentation will provide commentary on the response of mosquito fauna to changes in estuarine and freshwater habitats influenced by the Sydney Olympic Park urban development.

Using vector-borne disease outbreaks to guide ecosystem interventions.

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There is close association between ecosystem health and human health, exemplified by disease outbreaks that result from ecosystem disruptions. Historical examples include the massive mortality from yellow fever and malaria during construction of the Panama Canal, and more recently the emergence of Lyme disease in ecosystems with decreased biodiversity. In order to decrease the burden from such vector-borne diseases, it is necessary to identify disrupted ecosystems for early remediation. Human health is generally subject to the most widespread and comprehensive surveillance compared to other proposed 'indicators of ecosystem health'. Thus we suggest that human disease outbreaks are one of the most useful and practical bioindicators of the health of an ecosystem. Indeed, many subtle ecosystem disruptions are often only identified as a result of detailed epidemiological investigations following an anomalous increase in human disease incidence detected by routine surveillance mechanisms. Outbreak data can act as a pivotal warning system for ecosystem disruption and can be used to identify interventions for preservation of ecological (and ultimately human) health. Using this approach, appropriate strategies for intervention and remediation can be introduced at an earlier stage than would be possible based solely on environmental monitoring, thereby improving ecosystem health and human health concurrently.

Genotyping human and other vertebrate DNA from maggot gut contents: practical considerations

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Genetic analysis of maggot gut contents may aid a death investigator. We performed experiments aimed at defining procedures and conditions that will allow such an analysis to succeed. Calliphorid larvae were fed on beef, pork, rat, dog, cat, sheep, rabbit, deer, and human tissues. Larvae were then harvested at a range of developmental stages. Conventional forensic DNA typing procedures were performed on larval gut DNA extracts. The effects of maggot development, specimen preservation technique, larval starvation, and transfer of a feeding larva to a new food type, on recovery of the correct food genotype were examined. PCR amplification involved new cytochrome b primers specifically for vertebrate and not insect DNA, and a commercial STR kit. 20% bleach destroyed surface contamination without interfering with the analysis of internal tissues. Larvae manually removed from food or naturally postfeeding were unsuitable for this analysis after 24 hr off the food. For larvae transferred between food types, both the original and second food genotype could be recovered 46 hr following transfer if specific primers were used. Our methods failed with formalin-preserved specimens. However for all other preservation strategies STR and mtDNA types were obtained from at least some specimens after six months of storage.

Cellular and molecular immunology of the tick-host interface: implications for pathogen transmission

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Complex immunologic interactions occur at the tick-host interface. Tick feeding stimulates host innate and specific acquired immune responses to limit blood meal acquisition. In response to host defenses, tick saliva contains molecules that counteract host immunity. Tick infestation suppresses lymphocyte proliferation, polarizes/modulates cytokine responses, reduces adhesion molecule/integrin expression, modifies antigen presentation, and inhibits cytokine action. The cellular and molecular bases for these phenomena are being defined and multiple diverse pathways for achieving the same end point of immunomodulation are being characterized. The earliest stages of specific T-lymphocyte responses are being analyzed. Genomic and proteomic approaches are used to identify the molecules responsible for modulation of host immune defenses. Tick salivary gland gene expression is being characterized during the first few hours of feeding, and salivary gland molecules of interest are being cloned, expressed and their biological activities identified. Emerging from these studies is a picture of a web of interactions essential for successful blood feeding and transmission of infectious agents that is far more complex than initially envisioned.

Genomics and the development of vaccines against ticks and tick borne disease

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Whole genome sequences of major tick borne disease organisms are available or in progress. Sequencing of the first tick genome will commence soon. Host microarrays allow the dissection of the response to tick or pathogen infestation in a novel way. However, analysis of this deluge of information remains problematic. The rate-limiting step in vaccine development typically remains the assessment of candidate antigens and delivery systems in animals. Genomic information to be useful must be filtered to select a small number of candidates from a plethora of choices. Experience with *Boophilus microplus,* other tick species as well as *Anaplasma marginale* suggests that few highly effective antigens have yet been discovered and the reason for the efficacy of those remains obscure. Other, often less efficacious antigens identified to date fall principally into a few biochemical classes while many other potential antigen classes remain unexplored. The effective use of genomic information will demand a greatly enhanced understanding of the biology of vaccination than is currently available.

Seasonal studies on the influence of clothing and wrapping on carcass decomposition and arthropod succession.

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Six pig (*Sus scrofa* L.) carcasses, three wrapped and clothed carcass and three wrapped and naked, were sampled i) daily ii) after five days and iii) after ten days. Two more carcasses, one naked and not wrapped, and one clothed but not wrapped were sampled daily as controls. Differences in decomposition between the clothed or wrapped carcasses included the distribution and size of maggot masses and significant delay in ying out of the carcasses for all seasons. The maggot masses were primarily dominated by *Chrysomya marginalis* (Wiedemann) and *C. albiceps* (Weidemann), in autumn and summer. *Chrysomya chloropyga* (Weidemann) and *Calliphora vicina* Robineau-Devoidy in winter, with *C. chloropyga* and *C. albiceps* in spring. There was no delay in oviposition except in winter where a five day delay occurred on the wrapped carcasses. Extremely high internal temperatures in the warmer seasons caused significant maggot mortality on the wrapped carcasses. The predation of *C. marginalis* maggots by *C. albiceps* maggots was observed in autumn. In summer, maggot masses of these species were observed in separate groups. The coleopteran community was dominated by *Thanathopilus micans* L. larvae, succeeded by *Dermestes maculatus* De Geer, larvae. Adult *D. maculatus* and *Necrobia rufipes* De Geer, were present on the carcasses throughout all seasons.

Ectoparasites and arthropod-borne parasites of brown rat, *Rattus norvegicus*, in Ahvaz, southwest Iran

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Arthropods play an important role in the transmission of infectious agents to human and animals, such as rodents. Ahvaz, southwest Iran, is encountered with high density of rodents, especially *R. norvegicus*. In this study, 72 *R.norvegicus* were captured from different sites of Ahvaz, using live traps. Their ectoparasites were collected and identified. Thin and thick blood smears were prepared. After dissection of rats, their different organs were examined for any endoparasites, applying specific techniques. In general ectoparasites comprised 2 species of flea, *Xenopsylla astia* and *X. nubica*, 1 species of louse, *Polyplax spinula* and 1 species of mite *Laelaps nuttalli*. Among endoparasites, 9 species utilize arthropods as vector or intermediate host in their indirect life cycles. These included 1 species of protozoan parasite, *Trypanosoma lewisi* and 8 species of helminth parasites. The potential risk of each of these identified species to the health of human and animals will be discussed.

Applications of GIS and remote sensing for surveillance and control of mosquito borne diseases

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The risk of transmission of mosquito borne diseases depends on the spatial distribution of vector species in relation to the distribution of human hosts and, in case of mosquito-borne zoonoses, the distribution of vertebrate reservoir hosts. These distributions and temporal changes in these spatial patterns depend on environmental and climate/weather conditions. Satellite imagery provides means to gather vast amount of landscape/landcover and meteorological data that are proximate determinants of vector distribution and risk of disease transmission. Geographical information systems allow for the integration of environmental, entomological, epidemiological and demographic data and provide a framework for surveillance and control on scales ranging from local health departments and mosquito control districts to national and international programs. Products of applications of these tools and quantitative analysis of spatial data include data management tools, disease risk maps, and decision making tools for location and time of interventions. Examples from studies in the U.S. Israel, Trinidad and Kenya will be presented, including malaria cases in relation to *Anopheles* larval sites in Trinidad and Israel, dengue and dengue haemorrhagic fever in Trinidad, eastern equine encephalitis outbreaks in Michigan, USA, and La Crosse encephalitis transmission and the effect of introduction of *Aedes albopictus* in Illinois, USA.

Novel mosquito traps: can they control moquitoes

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A diversity of commercial traps for mosquito control have become available during the past five years. Most are marketed to homeowners. Many models have been evaluated in a large outdoor screened enclosure against two species (*Aedes aegypti* and *Ochlerotatus taeniorhynchus*) of laboratory reared mosquitoes to determine their relative efficacy. Several models were selected for field testing against natural populations. Ongoing research is trying to answer the question: Can mosquitoes be trapped out? Different management strategies are being evaluated, e.g. control potential on isolated islands versus urban/suburban residential settings. In urban/residential settings traps are being evaluated as a protective barrier around a city block and in individual yards. Preliminary results indicate that protective trap barriers provide excellent control (>90%) on isolated islands, but only moderate (50% reduction) control in urban/suburban areas. A single trap in a single yard provided no measurable mosquito control.

The mode of action of deet, SS220, and Bayrepel® arthropod repellents against *Aedes aegypti*, *Anopheles stephensi*, and *Phlebotomus papatasi*.

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Laboratory bioassays were conducted with human subjects using six-celled K&D modules and the known arthropod repellents, N,N-diethyl-3-methylbenzamide (Deet), (1S, 2'S)-methylpiperidinyl-3-cyclohexene-1-carboxamide (SS220) and 2-(2-hydroxyethyl)-1-piperidine carboxylic acid 1-methylpropyl ester (Bayrepel) against *Aedes aegypti* (Linnaeus), *Anopheles stephensi* Liston mosquitoes and the sand fly, *Phlebotomus papatasi* which are vectors of human diseases. When five females of each species were simultaneously exposed to one half repellent-treated ($4.8 \times 10^{-2} \mu mol/cm^2$) and one half untreated skin beneath the opening of a 3x4 cm cell, they bit exclusively on untreated skin. When treated and untreated skin was covered with cloth to preclude tactile contact with skin surfaces, the insects bit only through cloth covering untreated skin. The tests proved that each species was exquisitely capable of avoiding the compounds in their close-range feeding behavior and this was accomplished by olfactory chemical sensing. When cloth, untreated and repellent treated, was placed over the skin all species avoided the compounds and fed through the cloth only where repellent was absent. In the presence of any the three repellents within a cell, biting responses were reduced by about 50% relative to controls. Thus, besides inhibiting feeding on repellent-treated skin, the compounds also exhibited spatial-repellent activity.

Key issues in advancing the sterile insect technique for African malaria vectors

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Pilot projects that assessed the feasibility of the sterile insect technique (SIT) against Anopheles mosquitoes were conducted in El Salvador in the 1970s. Though on a relatively small scale, these trials demonstrated that sterility could be induced in a wild mosquito population through releasing (chemo)sterilized males. Since then, major scientific advances merit a renewed look at SIT and its applicability in the African context. We describe the R&D phase currently underway in Sudan and Austria, to develop appropriate mass-rearing systems and sterilization procedures. The salient features of such releases, also in relation to the use of transgenic approaches, will be discussed. These include colonization and mass rearing, (genetic) sexing, mosquito fitness and release mechanisms.

The influence of burning on the decomposition and insect succession of pig carcasses.

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Recent casework raised the question whether burning has an influence on the decomposition and insect succession of bodies. This was investigated by dousing three pig carcasses with different volumes of petrol and burning them to different degrees (Slight = 1.5L Medium =5L, Heavily = 10L). The insect succession onto these carcasses was compared to a control carcass. The control carcass decomposed at the fastest rate with the heavily burnt carcass at the slowest rate. Adult insects comprised the following: At the control carcass Diptera (60.8%), Coleoptera (29.9%) and Hymenoptera (8.8%). At the slightly burnt carcass Coleoptera (55.8%), Diptera (38.6%) and Hymenoptera (5.4%). The dominant orders at the medium burnt carcass were Coleoptera (38.5%), Diptera (33.3%) and Hymenoptera (28%). At the heavily burnt carcass Coleoptera (51.4%) and Diptera (28.6%) were dominant. Hymenoptera (18.7%) did not have a direct influence on decomposition, since they functioned as predators. *Chrysomya albiceps* was the dominant Calliphoridae species at the control and medium burnt carcasses and *Chrysomya marginalis* at the slightly and heavily burnt carcass. Representative samples of larvae collected during decomposition revealed that *C. albiceps* larvae preferred to feed underneath the carcasses, with *C. marginalis* larvae feeding mostly inside and at the top of the carcasses.

The study of excito-repellency phenomenon of *Anopheles stephensi* exposed to nets impregnated with cyfluthrin

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The excito-repellency phenomenon is occurred in the vector population following the application of definite insecticides. Based on the field observation, in relation to residual spraying operation, the excito-repellency phenomenon may led to behaviouristic changes of the mosquitoes as diversion either to new hosts or other unsprayed shelters. This occurrence may lead to failure in control program. At these conditions, the mosquitoes leave the sprayed houses to either unsprayed or outdoor shelters before picking up the lethal dose. The result of this investigation will be useful for planning of the control program in the country. For the first time, the standard design of excito-repellency (ER) test box was made of aluminum. The laboratory test was carried out with cyfluthrin treated nets against *An.stephensi* (Beech strain). The result showed that the tendency of the mosquitoes to long resting on the netting of exposure chamber was decreased at high dosage of insecticide, and tried to leave the exposure chamber toward the exit trap. The mean of the feeding rate was least at higher dosage in comparison of lower one.

Human behaviors and environmental factors elevating the risk of exposure to *Ixodes pacificus* nymphs in a hardwood forest

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The nymph of the western black-legged tick, *Ixodes pacificus*, has been implicated as the primary vector of the Lyme disease spirochete, *Borrelia burgdorferi*, in northwestern California. In 2002 and 2003, six human behaviors and environmental factors were evaluated as potential risk factors for acquiring nymphal ticks in a dense woodland in this region. The prevalence of *B. burgdorferi* and the causative agents of human anaplasmosis (*Anaplasma phagocytophilum*) and human monocytic ehrlichiosis (*Ehrlichia chaffeensis*) also were determined in nymphs that had been collected from subjects or by agging leaf litter. Time-adjusted tick-acquisition rates demonstrated that sitting on logs was the riskiest behavior followed, in descending rank, by gathering wood, sitting against trees, walking, stirring and sitting on leaf litter, and sitting on litter. The number of ticks acquired appeared to be unrelated to the type of footwear worn. Nymphal ticks were active throughout the daytime over a wide range of micrometeorologic conditions. Overall, about 3-4% of nymphs were infected with *A. phagocytophilum* and *B. burgdorferi*, regardless of the collection method, versus none with *E. chaffeensis*. Co-infection with *B. burgdorferi* and *A. phagocytophilum* was detected for the first time in this life stage of *I. pacificus*. The public health implications of these findings will be discussed briefly.

Genetically modified mosquitoes: a real world view

GC Lanzaro ADDRESS

The concept of controlling mosquito-borne diseases via the development and implementation of genetically modified strains has been under serious consideration since the early 1990's. Although this idea has been met with great enthusiasim in some quarters there remains substantial skepticism in others. The general concept and progress toward the development of such a strategy will be presented. Numerous roadblocks to the development of a GM mosquito plan remain, including (i) lack of gene drivers, (ii) lack of robust effector molecules and mechanisms to manipulate their expression, (iii) poor efficiency of transgenesis, (iv), and (v) no consideration of logistical problems associated with implementation in the field. These will be presented and prospects for overcoming them discussed. Aspects of the ecology and population genetics of targeted species will be discussed as they relate to the successful release and establishment of GM mosquitoes in nature.

Reverse chemical ecology: chemical prospecting of mosquito attractants and repellents

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For a few decades, chemical ecologists have been utilizing insect antennae as the sensing element or biodetector, in electroantennogram (EAG) and gas chromatography-electroantennographic detection (GC-EAD) assays for the identification of minute amounts of pheromones and other semiochemicals. The "molecular" equivalent of these electrophysiology-based "short cut bioassays" is the screening of attractants and repellents based on their binding affinities to odorant-binding proteins (OBPs). OBPs assist hydrophobic semiochemicals to cross an aqueous barrier in the insect sensilla (sensillar lymph) and reach odorant receptors (ORs). Both OBPs and ORs contribute to the high specificity of the insect olfactory system by forming two layers of filters. Although binding per se does not necessarily imply a physiological function, test compounds that do not bind to OBPs can be eliminated from further behavioral studies as they will never "see" the receptors. Reverse chemical ecology is a protein-based approach that requires the full identification of OBPs, cDNA cloning, functional expression of OBPs, and binding assays. In this presentation, we will discuss the latest development of a NIH-funded project* aimed at the development of better attractants and repellents for *Culex* species, the principal vectors of West Nile Virus.* NIH-National Institute of Allergy and Infectious Diseases (1U01AI058267-01).

Developing methodologies for a fungal biopesticide against *Boophilus* microplus, the cattle tick

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The Queensland department of primary industries and fisheries has been investigating fungal biopesticides for livestock ectoparasites as part of a wider strategy to reduce the reliance upon chemicals in livestock industries. Fungal biopesticides have been available for the control of agricultural and horticultural pests for over thirty years and methodologies and accepted protocols for the development of these fungal biopesticides are now well established. Current work with the cattle tick has shown that livestock pests present additional challenges, some of which are due to the inability to conduct *in vitro* assays on obligate parasites, the cost of *in vivo* experiments using animals and the different environments the fungus must operate in when invading the parasitic and free-living phases. In addition, the development of formulation and application methods suitable for penetrating an animal's hair coat and delivering a lethal dose of spores to ticks present further challenges. Solutions to these problems have sometimes been obvious and straightforward but mostly have been arrived at slowly and painstakingly. We have concluded that the development of methodologies for fungal biopesticides of livestock ectoparasites is a multidisciplinary exercise requiring a thoughtful balance of appropriate *in vitro* bioassays with on-animal investigations.

Increased densities of vector mosquitoes in anthropogenic environments in the Kapiti region in New Zealand and potential ecological mechanisms as to why

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Landuse changes, including deforestation, agriculture and urbanisation, have coincided with increases in mosquito-borne diseases worldwide. Anthropogenic environmental changes can alter mosquito populations by modifying the characteristics of breeding habitats, but we still poorly understand the ecological mechanisms involved. Mosquito breeding habitats were sampled in native forest, pastureland and urbanland in the Kapiti region, New Zealand, and 17 environmental characteristics were measured of each. Higher immature mosquito densities were collected from mainly artificial containers in urbanland and pastureland compared to mainly ground pools in native forest. Nine environmental variables were significantly different between landuses but none of these were significantly related to mosquito density. Principal components analysis partitioned environmental data into seven ecological types. One ecological type was significantly related to both landuse and mosquito density, and positively associated with ammonia, bacteria and dissolved organic carbon (DOC), and negatively associated with nitrate and predatory damselfly larvae. The findings of this study support those from prior surveys in warmer climes of greater vector disease risk in anthropogenically-modified environments because of compromised ecosystem integrity. Unlike most previous field-based work, this study also suggests important mechanisms involved; pinpointing mainly bacteria, DOC, nitrate and damselfly larvae as acting in combination to regulate vector mosquito density.

The final word on the genetic, evolutionary and taxonomic relationship of *Pediculus humanus* and *P. capitis*.

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Body lice are thought to have diverged from head lice when humans began to wear clothes. This recent evolutionary divergence and incomplete lineage sorting has complicated assessments of whether head lice and body lice are separate species. We used microsatellite markers to test lice from hosts that were infested with head and body lice simultaneously, to see if the head and body lice had interbred. Our results showed that head and body lice had not interbred on a host. Assignment tests revealed that lice that had migrated among hosts always moved from head to head, and body to body, but never between heads and bodies. We conclude that head and body lice are separate species. We speculate that head lice are descended from the lice of neanderthals and modern humans, and that body lice are descended from the lice of modern humans that left Africa about 100,000 ya.

The search for natural repellents from human beings against mosquitoes

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Blood sucking insects are differentially attracted to human beings. This is likely to be caused by differences in the semiochemical profiles of individual human hosts. Despite substantial research into insect-human interactions, the responsible semiochemicals have not yet been identified. An explanation for this difference is that unattractive individuals either lack attractant chemical compounds or may produce compounds that 'mask' the activity of attractants. A special air entrainment technique was developed and used to collect volatile chemicals from human volunteers, and the extracts analysed by gas chromatography (GC), coupled GC-electroantennography (GC-EAG) and GC-mass spectrometry (GC-MS). For the first time electrophysiological responses of female yellow fever mosquitoes, *Aedes aegypti* to volatiles from air entrainment extracts were recorded, and a number of electrophysiologically active compounds were identified. Previous behavioural studies have confirmed the differential attractiveness of volunteers to biting insects. A quantitative and qualitative analysis of compounds within the extracts, from individuals known to be differentially attractive to biting insects, has revealed significant differences in chemical profiles. The identification of behaviourally active compounds could lead to new and improved control technologies, whereby 'masking' compounds could potentially be incorporated into new, safe and natural repellents against biting insect pests.

Anopheles arabiensis at the edge of its distribution in Northern State, Sudan

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Northern state, Sudan is mainly desert, rainfall is rare and most life is found close to the Nile. *Anopheles arabiensis* is the only anopheline and malaria vector found in the area in recent decades. It is present along the Nile downriver to about 200 km south of the Egyptian border, where poor terrain and control efforts prevent its spread further north. Initial results are presented from field and molecular studies to determine the population structure and dynamics of the species and related environmental factors. The receding Nile following the flood contributes most to sustaining *A. arabiensis*. Larval breeding sites can be found on low lying riverbank, ied-up inlets, or channels around islands, through most of the year. Land use is mainly associated with irrigation, but this does not appear to contribute greatly to anopheline numbers, however the extent of human presence and activities do increase the potential for anopheline breeding. Upriver from the fourth cataract and downriver from the third cataract, the terrain is rocky desert and less suitable for both humans and mosquitoes. The *An. arabiensis* population is thus semi-isolated within an approximately 700 km corridor a few kilometres wide and breaks down further into pockets depending on particular river features.

Control of biting midge (Ceratopogonidae) attack on residents of a marine canal estate in Redcliffe (Australia) by residual barrier treatment of Bistar 80SC® to ornamental garden plants in house yards

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Biting midge (Ceratopogonidae) breeding in marine sediments associated with engineered canals in a residential community in Redcliffe Australia have been responsible for considerable biting attack and nuisance. Limitations on approved applications of larvicides in breeding sites has historically restricted control to personal protection, midge screening and education of residents. In October 2003 – Feb 04, Redcliffe City Council funded a trial to test the efficacy of Bistar 80SC® (FMC Chemicals Pty Ltd) as a 0.1% AI (Bifenthrin) residual application to ornamental garden vegetation and dwelling external walls. Eight paired randomised treated and untreated premises were assessed for differences in biting midge exposure. The assessment was undertaken using CO_2 and Octenol baited light traps placed in treated and untreated yards. A self administered questionnaire scoring individual household daily experience of biting midge activity was also used. Untreated control premises received a placebo application of water alone to control bias in the resident questionnaire. Analysis of difference in light trap collections between treated and untreated showed a 76.4% reduction in midge (primarily *Culicoides molestus*) from treated premises. Resident survey results indicated occupants of treated premises experienced generally favourable biting midge conditions while residents from untreated homes experienced fluctuating exposure and discomfort corresponding to lunar emergence cycles.

Aerial efficacy assessment of Anvil 10+10 against *Anopheles quaimaculatus*

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Aerial ULV applications of Anvil 10+10 were conducted against caged, field-collected *Anopheles quaimaculatus* adults using Beecomist® 360A rotary atomizers. Standard and optimized applications were conducted at 0.62 fl oz (0.0036 lbs a.i.) per acre using stacked passes 3,500 ft and 5,000 ft upwind of the target site. Efficacy of Anvil 10+10 and impact of wind velocity at flight altitude (150 ft) were assessed by bioassay and analysis of collected droplets.

The potential role of aquatic insects in the transmission of Buruli ulcer disease in Africa

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Buruli ulcer is the third most frequent mycobacterial disease in humans, next to tuberculosis and leprosy, often causing serious skin ulcerations, bone deformities, and disability. Unlike TB and leprosy, Buruli ulcer has emerged amatically since the 1980's, with increasing numbers of cases being reported from at least 31 countries, mostly in tropical and subtropical areas, including Africa. Although poorly understood, the epidemiology of Buruli ulcer associates the resurgence with people who live and/or work close to wetlands, especially slow flowing (riverine) or stagnant water bodies (marshes, swamps), often created as a result of some human environmental disturbance. The disease is not known to be contagious between humans, and epidemiological studies have not yet established natural reservoirs or modes of transmission. However, it was recently demonstrated experimentally that *M. ulcerans* could survive and multiply exclusively within salivary glands of aquatic bugs (Naucoridae: *Naucoris* sp.) and they were able to transmit *M. ulcerans* to mice by their bites, suggesting a possible mechanism for human infection. The organism has been found in samples of water, biofilm and detritus. A possible trophic scenario for the natural mode of transmission of *M. ulcerans* is presented based on preliminary work in Ghana by the authors.

Efficacy of a Spinosad suspension concentrate formulation for controlling chewing and sucking lice on cattle under field conditions

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Field studies were conducted with cattle in Wisconsin and Illinois between January and April, 2000, to determine the efficacy of 0.04% spray and 2 mg/kg pour-on treatments of a spinosad suspension concentrate formulation against chewing and sucking lice. Positive control substances included in the study were Co-Ral® EC (5.8% coumaphos) and CyLence™ Pour-on (1% cyfluthrin). Animals in the negative control group remained untreated. All lice species were visually sampled on each animal at weekly intervals after treatment, for eight weeks. Individual louse counts were summed over each examination site for each species. A repeated measures analysis using a mixed model (SAS PROC MIXED), was conducted for data pooled across both study sites. Both spinosad treatments and CyLence™ Pour-on provided excellent efficacy (98-100%) against *Bovicola bovis* over the entire study period; Co-Ral® EC efficacy opped below 90% from Weeks 6-8. The efficacy of the spinosad spray treatment against *Linognathus vituli* was >93% for the entire study and >88% for the spinosad pour-on treatment. Co-Ral® EC was >90% efficacious against *L. vituli* during Weeks 1-7, while CyLence™ Pour-on only exceeded 90% for Weeks 3-7. *Solenopotes capillatus* were found only at the Wisconsin site; all treatments were highly efficacious throughout the study period.

The effect of human age on Dengue seroconversion risk in Iquitos, Peru

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Basic models of infectious disease often assume that force of infection is independent of host age. This assumption has been shown to be violated for many directly transmitted diseases, but has rarely been examined in vector-borne systems, such as dengue. Data from DNA fingerprinting studies of mosquito bloodmeals indicate that the biting rate of *Aedes aegypti* increases with human age. If risk for dengue infection increases with human age, models assuming an age-independent risk will underestimate the proportion of the population that needs to be vaccinated in order to eliminate disease transmission. A cohort of approximately 3,000 schoolchilen and their families in Iquitos, Peru were followed for up to 5 years (1999-2003) with blood samples collected at 6-month intervals. Sera were tested for dengue virus antibodies by the plaque reduction neutralization test. The effect of human age on risk for dengue seroconversion was evaluated using a multiple logistic regression model, which also included area of residence within the city. Preliminary analysis suggests that risk increased with age and was highest in the southern region of the city. Estimates of age-specific forces of infection can be used to design an optimal immunization program.

Oviposition site selection by mosquitoes in response to drought

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Mosquitoes are vectors of many important medical and veterinary diseases. Although outbreaks of mosquito borne disease can be correlated to changes in precipitation in the very short term, precipitation rarely corresponds to mosquito abundance in the long term. Here, I present the results of a field based mesocosm experiment designed to test how water level and drought influence the oviposition behaviour of two species of mosquitoes. I used two water levels (high and low) and two drought treatments (normal and drought). Water for the drought treatments was obtained from ponds that had experienced drying for a period of a month prior to experimentation. The two mosquito species differed markedly in their oviposition response. The egg laying behavior of *Ochlerlatatus notoscriptus* was significantly greater in the drought treatments regardless of water level. However, the egg laying behavior *Anopheles annulipes* showed the opposite response with significantly more larvae present in the high water treatments regardless of whether the treatment water was normal or in drought. My results provide supportive evidence that mosquitoes may show population outbreaks following drought years and that this framework may provide a reliable way of predicting year-to–year variation in mosquito abundances based on individual species oviposition response.

Tests on a new bait for strike flies (Diptera: Calliphoridae)

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A bait consisting of sulphur-containing volatile compounds encapsulated in a caesin-based slow release matrix was tested for its efficacy in catching sheep strike flies (Diptera, Calliphoridae). The fresh bait was significantly more effective than a liver/sodium sulphide mixture in trapping a major strike fly (*Lucilia sericata*) and a minor strike fly (*Chrysomya rufifacies*) on a sticky trap. The bait also trapped significantly more gravid flies of these species. The bait efficacy wore off after 17 days under ambient conditions. The bait was also less effective than the liver mixture when used in Western Australia traps. This may be due either to its loss of effectiveness with age, or to differences in the responses of flies when landing close to the bait on a sticky trap and when searching for the exact location of an odour source in a Western Australia trap. Ways of improving the bait efficacy are discussed.

Aedes aegypti surveillance in Iquitos, Peru: a 4- year test of the pupal/demographic survey

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We present results from 4 years of pupal/demographic surveys of a circuit of ≈ 6,000 houses surveyed >10 times in the Amazonian city of Iquitos, Peru. We compare traditional larval indices with pupal and adult mosquito indices. We evaluated *Aedes aegypti* production from different container classes, including those based on water management practices overtime and space. A total of 289,941 water-holding containers were characterized, of which 7.3% were positive for *Ae. aegypti*. Although, temporal and geographic variations were detected for all variables examined, consistent production patterns were observed for the lid status (lids: 32% wet containers, 2% pupal production), container location (outdoor: 43% wet containers; 85% pupal production) and method by which the container was filled with water (rain filled: 15% wet containers; 88.3% pupal production). Based on these patterns and additional targeting of containers with highly variable pupal counts we propose a targeted control strategy for Iquitos that would reduce *Ae. aegypti* populations below published but untested threshold values for Iquitos.

Screening medicinal plant extracts for effects on *Aedes aegypti* (Diptera: Culicidae) & toxicity to a non target organism

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A preliminary study was conducted to find out the effects of the extracts of 112 medicinal plant species, collected from the southern part of Thailand, on *Aedes aegypti*. Studies on larvicidal properties of plant extracts against the third and fourth instar larvae of *Ae. aegypti* showed fourteen species with high toxicity. Six plant species were comparatively more effective against the larvae at very low concentration. These extracts demonstrated no or very low toxicity to guppy fish (*Poecilia reticulata*). Three plants with promising larvicidal activity, *Mammea siamensis*, *Anethum graveolens* and *Annona muricata,* were used to study of the impact of the extracts on the life cycle of *Ae. aegypti*. The results showed that these plants affected the reproductive potential of surviving adults by reducing the number of eggs laid and egg hatchability. The first instar larvae were very susceptible to *A. muricata* and the second instar larvae were susceptible to *A. graveolens* while the third and fourth instar larvae were susceptible to *M. siamensis*. These extracts delayed larval development and inhibited adult emergence.

The mosquito burden in Kenya

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Kenya is a tropical country with a climate favourable for diverse insect life – both the harmful and the useful alike. Embu District experienced a malaria outbreak between January and February 2001 where more than 300 people were admitted at the main health facility in the District, 70 lives were lost. Malaria is endemic in Embu district with a morbidity of 60% and mortality of 25%. Expectant mothers and chilen are affected most with slide positivity of 30% and 45% respectively. Annually, each household spends around US$ 65 on malaria control and funeral expenses. Larval densities range from 30-40% with peak densities in March, April and May. Despite this, 25% of the community cannot positively identify mosquito as the malaria vector. Only 12% can correctly name the five main symptoms of malaria (fever, chill, sweating, anorexia, joint pains). The ministry of health has issued guidelines on uniform treatment and community sensitisation. Consistent use of bednets, chemoprophylaxis and indoor residual spraying are being encouraged. Plans are underway to start biological adult and larval control. This paper discusses the mosquito burden from the perspectives of larval density, host prevalence index, mortalities and morbidities, bed occupancy and the economics of vector control

Tick vector-pathogen interrelationships

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The interrelationships between tick-borne pathogens, their tick vectors and diverse vertebrate hosts, are represented by a triangle of parasitic interactions. The interactions are between (i) tick-pathogen, (ii) pathogen-host, and (iii) tick-host. In (i) the pathogen interacts with its vector, infecting and replicating within tick cells or extracellular spaces. The ability of a particular tick species to act as a vector depends on whether the pathogen can survive and overcome several 'barriers' within the tick. However, this is a simplistic view of the tick vector-pathogen interrelationship as both (ii) and (iii) influence the success of (i). This triangle of interactions is focused at the skin site of tick feeding - the initial site at which pathogens gain access to either their host or vector. For pathogens, the skin-feeding site is an ecologically privileged niche, profoundly modified by the pharmacological activities of bioactive tick saliva molecules. Exploitation of this niche is known as saliva-activated transmission (SAT). Tick saliva molecules that promote SAT have not been identified but appear to differ for different tick vector-pathogen interrelationships. Identifying the SAT factors and their mode of action offers new insights for controlling ticks and tick-borne diseases.

Argentina: veterinary forensic entomology as a potential tool against rustling

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Cattle rustling entails economic loss to cattle owners and danger to their safety, marketing of uninspected meat and hides and loss to the State from unpaid taxes. In 2003, the Faculty of Veterinary Medicine (Tandil, Buenos Aires province, Argentina) incorporated forensic entomology to a course of legal veterinary medicine aimed at fighting rustling. Preliminary field experiments showed a large proportion of hemisynanthropic insects, most of them Neotropical, which may represent the autochthonous necrophagous fauna, now displaced by synanthropic introduced species in urban environments. Colonization and development times proved longer in the arid climate of Tandil than at Buenos Aires city, where most published data were gathered. Field experiments with bovine bodies and parts are required. Hitherto, bovines have been avoided as a model because of the added complication of the ruminant digestive tract. Since most countries with rustling problems also have a pattern of introduced species displacing the original necrophagous fauna in city environments, they might benefit from knowledge obtained through these studies.

Arboviruses, mosquitoes & rNA interference

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The identification of critical arbovirus-vector interactions reveals new insights into vector competence. For instance, flavivirus and alphavirus replication in mosquitoes triggers a natural RNA interference (RNAi) response; however, viral counterdefenses probably allow RNAi evasion and establishment of persistent infections. We can enhance RNAi to dengue virus type 2 (DENV2) in *Aedes* species by administration of double-stranded (ds) RNA derived from the DENV2 genome, either by direct transfection of dsRNA, transient expression from a Sindbis virus (SINV) transducing system, or stable transformation with a plasmid that expresses a hairpin-forming transcript. RNAi is sequence-specific, is maintained by low concentrations of effector RNA, blocks accumulation of intracellular viral RNA, and results in the appearance of 21-25 nt siRNA homologous to the mRNA target. We have identified genes from the Dicer and Argonaute families in 2 mosquito genera and have used RNAi to knock down their expression to determine which genes have a role in virus resistance. We have evidence that DENV2 and some strains of SINV establish persistent infections in mosquitoes by suppression of RNAi, and are investigating mechanisms. We are identifying viral-encoded suppressors by site-specific mutations in infectious cDNA clones. Our work suggests that persistent infections in mosquitoes result from a balance between host RNA silencing and viral counterdefenses and provides new strategies for tipping the balance in favor of the vector host.

Endosymbionts & sex ratios in the human head louse

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At the turn of the 19th century the first observations of a female-biased sex ratio in broods and populations of head lice had been reported. A study by Buxton in 1938 on the sex ratio of lice on prisoners in Ceylon is still today the subject of reanalyses. However, in the last sixty years no data have been collected, especially on more normal levels of infestations. Here we report a female bias of head lice in a survey of 480 school children in Argentina. This bias is independent of lice intensity. These lice as well as lice from pupils in Britain were carrying several, related strains of the endosymbiotic bacterium *Wolbachia*, one of the increasing number of intracellular sex ratio distorters. No lice without *Wolbachia* were detected. Similar *Wolbachia* strains are also present in the pig louse suggesting that this endosymbiont might have marked influence on the biology of the whole order. The sex ratio distortion in head lice that has been reported world wide is stable over time; that is a remarkable deviation from the stability of frequency-dependent selection of Fisher's sex ratio.

Blood feeding in *Cnephia* (Diptera: Simuliidae): molecular phylogeny

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Five species of *Cnephia* were studied, three non-biting, one a mammal feeder, and one a bird feeder. Of the non-feeding species, *C. eremite* is Holarctic in distribution and specimens were examined from both Canada and Sweden. *Cnephia pallipes* is a Palearctic non-feeder and *C. dacotensis* a temperate Nearctic non-feeder. *Cnephia pecuarum* is an economically important large mammal feeder in south-central U.S. and *C. ornithophilia* a widely distributed bird feeder in southern and eastern North America. Sequence was obtained from the complete mitochondrial CO-II gene (687 bp) and partial EF1-alpha nuclear gene (1030 bp). Molecular data is discussed in relation to conventional wisdom about relationships among species based on cytological and biological information, geographic distribution and genetic diversity within species. Molecular data strongly support *Cnephia* as a monophyletic clade in the tribe Simuliini. Speciation hypotheses were not fully concordant between molecular and conventional data.

Onchocerciasis in Central America: ecology & control

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Detection of O. volvulus larvae in vector populations of black flies is of prime importance in the assessment of the effectiveness of ivermectin control programs. Detection of larvae is attained by dissection of individual flies or by PCR (amplification of an Onchocerca 150 bp repeated sequence, followed by hybridization of the PCR products with a species-specific DNA probe) from pools of flies. The PCR was compared with dissection method to estimate the infection rate (p) and the mean larval load (any larval stage) per parous fly (m) in a local vector population. Both methods produced statistically similar estimates, thus PCR is an alternative and efficient method for entomological surveillance. Sample sizes required to estimate m from p for fixed levels of precision were calculated using Mexican and Guatemalan data, assuming that k (an aggregation parameter from the larval distribution among parous flies) is known. Optimal sampling and PCR was used to evaluate the impact of ivermectin on m, p and transmission potentials in Mexico. The data suggest that substantial progress towards the goal of suppressing transmission has been accomplished. In southern Chiapas, entomological estimates decreased in comparison with the baseline data, but PCR positivity was still detectable. In northern Chiapas and Oaxaca, with no baseline data, there was an absence or near absence of infected and infective flies.

Malaria vectors, *Anopheles hyrcanus* group (Culicidae, Diptera): morphological comparisons & molecular analysis

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Anopheles hyrcanus group consists of at least 30 species, with several species including *Anopheles sinensis, lesteri, pullus*, that are known or potential malaria vectors. The morphological diagnostic features and molecular identities of malaria vector *hyrcanus* group species are determined. Larvae, pupae and adults of selected species were described morphologically, and their ITS2 and DNA sequences from our laboratory and GenBank were compared.

Distribution & bionomics of malaria mosquito vectors, *Anopheles (Anopheles)* in Japan, Korea & other parts of Asia

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Mosquito specimens of *Anopheles (Anopheles)* from collections conducted in Japan (2002-03), Korea (1999-2003), Philippines (2002), HongKong (2001), mainland China (2001-03) were identified using morphological and molecular techniques, and recorded. Larval, pupal and adult rearings of field collected samples were conducted for morphological and molecular analysis. Detailed locations of various collection sites and habitats were characterized for known and potential malaria vectors of different countries. Habitats of vectors include ricefields, irrigated ditches, temporary and permanent pools, ponds, streams, others.

Dengue mosquito vectors, *Aedes* and *Ochlerotatus*: morphological identifications & distribution

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The diagnostic morphological features of mosquito vectors of dengue viruses are compared for various *Aedes (Diceromyia, Stegomyia)* and *Ochlerotatus (Finlaya, Gymnometopa)* species found in four regions of the world, namely Oriental Region, Pacific and Australian Region, Afrotropical Region, and the Americas. Morphological characters are indicated to separate both larvae and adults of the vector species of each region. The distribution and bionomics of *Aedes* and *Ochlerotatus* vector species are included.

New & resurgent arboviral disease threatening humans in Australia

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Australia has a range of indigenous arboviruses and an introduced few that are of public health concern; all are mosquito-borne. Endemic encephalitogenic viruses (Murray Valley Encephalitis, Kunjin) are annually active and responsible for occasional cases in the northwest but, despite widespread vector distribution, only rare activity and no epidemics in the southeast since 1974. The most common arboviruses, Ross River annually infecting many thousands nation-wide causing polyarthritis, and Barmah Forest (a similar syndrome) on the increase and responsible for outbreaks of hundreds regionally, have a spectrum of coastal saline and inland freshwater vectors. The most important exotics are the returning dengue viruses with outbreaks of hundreds confined to Queensland by vector distribution but with virus endemicity and vector spread threatening, and the recently introduced Japanese Encephalitis virus causing only a few deaths in northern Queensland but with virus persistence and its affinity for a local vector threatening. Environmental factors drive arbovirus activity by influencing vector populations, which vary with virus and circumstance, although little is known of the role of reservoir hosts. Tourism and development activities that increase human/vector/virus contact, and climate changes that promote vector and virus proliferation, threaten to exacerbate the disease threat to the public health.

Repellents for integrated tsetse management

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ICIPE is in the forefront in identifying potent repellents, both synthetic analogues of natural tsetse repellents and natural repellent blends from un-preferred animals like waterbuck and zebras. One potent synthetic repellent for savannah tsetse species, developed by molecular optimization studies, acts as an olfactory antagonist of a key phenolic kairomone, which flies use to locate the host for feeding (Saini & Hassanali; patent application). This repellent substantially reduce tsetse challenge (> 80%) and feeding efficiency (>80%) on cattle. A prototype dispenser which is placed around the neck of an animal has been developed. Field trials indicate that the repellent can provide effective protection to cattle but all animals in a herd need to be protected or else significantly higher infection occurs in unprotected animals. Experiments are ongoing to evaluate how repellents can be integrated with other existing tsetse control techniques. A 'push-pull' strategy that deploys repellents (to 'push') and baited traps / targets (to 'pull') to enhance suppression rates is being evaluated. A variant of this may involve the use of a proportion of cattle with 'pour on' insecticides as live baits. A business plan for commercial production and distribution of repellent products is also being developed.

Mitochondrial genome organization of lice is highly variable

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Mitochondrial (mt) genome organization is extremely conserved in mammals, vertebrates and most arthropods, e.g., humans and frogs have the same mt genome organization; so have fruit flies and water fleas. We have sequenced the entire mt genomes of the wallaby louse (Phthiraptera: Amblycera) and the pigeon louse (Ischnocera), and part of the mt genomes of the human louse (Anoplura) and four other amblyceran lice. Unlike most arthropods, the mt genome organization of these lice is highly variable. Lice share only 1-2 gene boundaries with fruit flies. No gene boundaries were shared among lice of the three suborders. Mt genome organization differs even among three genera within the suborder Amblycera. Changes in the mt genome organization of lice include gene translocation, inversion, separation of rRNA genes and loss or multiplication of non-coding regions. In addition to lice, we found mt genome organization was variable or highly variable in other two hemipteroid orders, Psocoptera (psocids) and Thysanoptera (thrips), but is strictly conserved in the order Hemiptera (bugs). We speculate that the changes in the mt genome organization in lice, psocids and thrips may be related to their life-history traits such as small body size, small colony size, and high degree of inbreeding.

Impact analysis of certain phyto-extracts on the life cycle of malaria vector

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The petroleum ether extract (Pe. E.) of *Artemisia annua* and *Azardirachta indica* have remarkable influence on the lifecycle of malaria vector, *Anopheles stephensi* by arresting larval and pupal period. These phyto-extracts hamper the hatching of larvae and further deform the larvae. The retarded emergence of adults was observed due to percentage decrease in transformation of eggs into larvae from 100 to 75% and pupal from 80 to 45% at concentration variable from 7.50 to 240 ppm of Pe. E. of *A. annua* and 80 to 45% and 79 to 66% at concentration range from 6.25 to 100 ppm of Met. E. of *Az. indica.* The percent larval & pupal mortality range was observed from 20 to 55 and 19 to 83.50% on exposure to *A. annua* extract and 21 to 34% and 27 to 84% on treating with *Az. Indica* extract. The decreasing trend in the growth index was observed in both the extracts. These phyto-extracts have great potency and may be recommended as economical, ecofriendly and effective tool of bio-control of this highly nuisance vector.

Bidirectional signalling and synaptogenesis: semaphorin1a and the transition from growth cone to synapse.

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Target recognition during which a motile growth cone is transformed to a sedentary synapse is poorly characterized at the molecular level. We have demonstrated in the *Drosophila* giant fiber system that this transition is regulated in part by semaphorin1a (Godenschwege et al., 2003). Sema1a is a trans-membrane member of a family of proteins typically thought of as repulsive ligands that guide growth cone path finding. We have shown that Sema1a is required in both the pre- and postsynaptic cells for normal synapse formation and are using Sema1a as a point of entry to discover the signaling systems that regulate the transition from growth cone to synapse. In the pre-synaptic giant fiber Sema1a functions as a repulsive receptor and when Sema1a is over-expressed in the giant fiber synapse formation is disrupted. This gain-of-function defect can be enhanced by over expression of rac1 and suppressed by expression of the dominant negative rac1 suggesting that the repulsive function of sema1a is dependent on the small GTPases. We will elaborate on the role of rac1 as well other signaling systems that interact with semaphorin1a and are required for synaptogenesis . Finally, a model for the assembly of this giant synapse will be presented.

Adult survival and reproductive effects of forced egg-retention time in *Aedes albopictus* after application of deet as an oviposition deterrent

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The insect repellent deet (0.1% concentration) used as mosquito oviposition deterrent in the laboratory could influence the gravid female *Aedes albopictus* Skuse to retain and maintain mature eggs in her body. The egg-retention in female mosquitoes could benefit survival of the gravid females. The gravid females can lay the maintained eggs on availability of water or another liquid medium in the cage, but the length of forced egg-retention time could reduce the mean number of eggs laid per female. The gravid females that retained eggs also laid a higher percentage of non-tanning eggs, which increased with the egg-retention time. The rate (%) of egg hatch was not affected by deet used as oviposition deterrent. It was however, affected by egg-retention time in both treated (exposed to deet) and untreated (control) gravid females. The rate of egg hatch reduced after 3 weeks of retention, and after 6 weeks of retention the eggs of both treated and control gravid females could not hatch. The fecundity and fertility of gravid female *Ae. albopictus* was affected by the time duration of forced egg-retention.

Haematobia irritans (Diptera: Muscidae) mitochondrial genome amplification and sequencing: potential of the long-PCR approach for calyptratae mitogenomics

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Long-PCR strategies have significantly improved the sequencing and characterization of insect mitogenomes essentially by providing an alternative way to purify sufficient mtDNA from limited amounts of individual template and thus stimulating a wide range of otherwise limited techniques on insect molecular and evolutionary biology. We optimized Long-PCR reactions for the amplification and sequencing of the complete mitogenome of the horn-fly, *Haematobia irritans* (Diptera: Muscidae), a key parasitic fly of cattle. *H. irritans* mitogenome was amplified using four conserved Long-PCR primers designed on Dipteran 16S lrDNA gene combined with two previously described 'universal' primers. Amplified products ranged from 8.5 to 9.3Kb depending on primer combination, with a 1.5Kb overlap. We obtained specific products on a microgram range providing sufficient mtDNA for several techniques, including RFLP analysis and the sequencing of the *Haematobia irritans* mitogenome from a shot-gun library approach. Using H16S primers, we also amplified long-PCR products from seven other Calyptratae key species of medical-veterinary importance: the screw-worm fly, *Cochlyomyia hominivorax*, the blowflies *Chrysomya albiceps*, *Chrysomya megacephala*, *Chrysomya putoria*, *Lucilia cuprina*, the botfly, *Dermatobia hominis* and another muscid, *Musca domestica*. These results contribute not only to achieve complete *Haematobia irritans* mitogenome but also stimulate Calyptratae mitogenomics.

The complete mitochondrial genome of the human botfly *Dermatobia hominis* (Diptera: Oestridae)

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"

The botfly *Dermatobia hominis* is a parasite of humans and livestock, producing a furuncular myiasis. The larvae of *D. hominis* develops in the subcutaneous tissue causing economic loss and health problems, being one of the most important livestock pests in Latin America. The complete mitochondrial DNA (mtDNA) sequence of *D. hominis* was determined and has 16360 bp. The nucleotide composition is biased towards adenine and thymine, which accounted for 77.8% of the whole genome. The *cox1* gene has a Serine as the start codon, while incomplete termination codons were found for *cox1*, *cox2* and *nd5*. The control region, protein-coding genes, rRNA and tRNA genes are in the same order and orientation when compared to other dipteran mitochondrial genomes, except for a duplication of $tRNA^{Val}$ located approximately in the same position of the $tRNA^{Asp}$, but in the opposite strand. The identification of restriction sites on the sequenced mtDNA and the correlation with previous RFLP analyses provided insights for an overall scenario of the genetic variability and population structure of *D. hominis*, being helpful for monitoring the variation and dynamics of this species. The availability of complete mitochondrial sequences is an important source of sequence information for Diptera molecular and evolutionary approaches.

Are *Pediculus humanus* infected with *Wolbachia* spp.?

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Wolbachia spp. belong to the alpha-subdivision of the Protobacteria and are closely related to the Rickettsia and their relatives (Class Alphaproteobacteria, Order Rickettsiales). *Wolbachia* spp. are obligate intracellular bacteria and have been recorded in approximately 76% of insects studied. *Wolbachia* spp. are transmitted from mother to offspring through the cytoplasm of the egg. Horizontal and accidental transmission have also been recorded. The impact of a *Wolbachia* sp. on its host may vary. Enhanced fertility or fecundity, pathenogenicity, parthenogensis, feminisation, male-killing as well as cytoplasmic incapability (unidirection and/or bidirectional) may be caused by *Wolbachia* spp. infections. Cytoplasmic incapability has been linked to the transmission and inheritance of mitochondrial DNA in that it may provide the impetus for selective sweeps. The sweep of a mitochondrial-type through a population and from generation to generation may be enhanced in lice due to the short generation time, small population size and the founder effect that occurs as a result of dispersal of lice. Here we present our data on whether or not *Pediculus humanus* (body/cloths louse) and *Pediculus capitis* (head lice) may be infected with *Wolbachia* spp.

Triatoma mexicana in the state of Guanajuato, Mexico: preliminary results.

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Mexico have 7 genera and 24 species of Triatoma genus which are *Trypanosoma cruzi* vector's, some with habits intradomicile. In Guanajuato state have been reported 5 triatomines species, among them *Triatoma mexicana*, which is little studied. With the purpose of know their ecotopes, distribution, infection for *T. cruzi*, morfometric, citogenetic and of life cycle in the laboratory, 9 localities of three municipalities were studied in the northeast of the State. We collected 110 triatomines, adults 91(83%), nymphs 19 (17%), males 80 (73 %) and females 11 (10 %). Positive to *T. cruzi*. 5 (4.54%). Capture sites, 6 (4%) were captured indoors and 104 (96%) in the outdoors. The main ecotopes in the intradomicile were: bed, wall, kitchen light bulb and in the peridomicile were: external wall, courtyard light bulbs, stone wall and farmyard. This specie is attracted by the artificial light (tungsten). The season with more triatomines was spring. Studies of morfometric , citogenetic and biological cycle are not concluding. This vector doesn't present intradomiciliated habits like initially it had been reported, their biological cycle performance in the peridomicile is in the stone wall and farmyard in these sites we captured the biggest number of triatomines in the nymphs phase

Vertebrate integumentary chemicals: endogenously produced and acquired chemicals use to deter arthropods

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Ectoparasitic arthropods may be deterred from feeding on certain vertebrates by substances endogenously produced in the skin, pelage or plumage. Deterrent chemicals may also be acquired by them by rubbing against exogenous sources, i. e. anointing. Monkeys and birds rub themselves with millipedes that defensively discharge benzoquinones. Laboratory bioassays showed that three benzoquinones in millipede secretions deterred ticks and mosquitoes by repellency, toxicity and behavior alteration.

Spatio-temporal analysis of reinfestation by *Triatoma infestans* (Hemiptera: Reduviidae) following insecticide spraying in a rural community in northwestern Argentina.

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The spatio-temporal reinfestation patterns by *Triatoma infestans* following a blanket insecticide spraying in a rural community (Amamá) was analyzed using geographic information system (GIS), satellite imagery, and spatial statistics. Domestic and peridomestic reinfestation by triatomine bugs was monitored from 1993 to 1997 after the apparent elimination of *T. infestans* in 1992. In addition to domiciles, *T. infestans* occupied pig and goat corrals, wood piles, trees, and additional sites associated with chickens. *T. infestans* was detected at least once in 75% of 2110 sites evaluated. The prevalence of sites positive at least once for *T. infestans* during the study period increased sharply from 1993-1995 (0.6-2.9%) to November 1997 (32%). The initial source of *T. infestans* was a pig corral in southern Amamá one year post-spraying. Subsequent infestations were clustered around this initial focus at a distance of ~400 m starting in 1995. In 1996, clustering was maximized in sites within the same compound or in neighboring compounds at distances of 25-175 m. These results indicate that an effective control program on the community level will be based on the spraying of actual epicenters as well as sites within a 450 m of these epicenters to prevent the propagation of *T. infestans*.

Vector biology and control research in southern Africa

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In collaboration with the World Health Organization Regional Office for Africa in Harare, Zimbabwe, a multicentre study for the characterization of vector populations and capacity building for vector control in seasonal, epidemic-prone malaria transmission areas of southern Africa, was initiated. Namibia, Botswana, Zimbabwe, Swaziland and South Africa participated in mosquito collections, insecticide susceptibility testing and involvement in malaria transmission. *Anopheles arabiensis* was the major malaria vector in all five countries. Only Zimbabwe and South Africa gave indications of insecticide resistance in *An. arabiensis. Anopheles merus* was implicated in malaria transmission at one locality in central Zimbabwe. A further study on insecticide resistance in northern Kwazulu/Natal Province of South Africa was undertaken. The efficacy of spraying houses with both DDT and pyrethroids, either together as a single mix or one on top of the other was investigated as a possible resistance management tool. Capacity building was a major component of both these projects. Collaborators processed their own samples in the laboratory in Johannesburg, and two were registered for Masters degrees at the University of the Witwatersrand.

Identification of gene candidates for age determination in *Aedes aegypti* (Diptera: Culicidae)

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Aedes aegypti is the principal vector of dengue, a disease of increasing global significance. Vector population age structure is a critical determinant of a vector population's capacity to transmit disease. Current age-grading methods are of limited value as age estimates are restricted to 15 days, while only females older than approximately 14 days can potentially transmit dengue. Development of an age-grading method that predicts mosquito age over the entire lifespan of *Ae. aegypti* would be a valuable tool for disease transmission studies and evaluating vector control strategies. Here, we describe gene candidates that show age related expression profiles, for a rapid PCR-based method of age determination. Candidate genes have been identified from published *Drosophila* microarray data of gene expression across adult age. *Drosophila* candidate gene expression was confirmed using Northern blots of pooled RNA samples across a range of age groups (adults 1-29 d, with 4 d interval). Gene orthologues from *Ae. aegypti* were isolated by PCR using degenerate primers and gene expression profiles were validated by Northern blots analysis as for *Drosophila*. The application of real-time reverse transcriptase PCR techniques to determine gene expression levels in individual female mosquitoes and the statistical tools to predict age will be presented.

Detection and characterization of the tRNA^{Ile} in the mtDNA control region of myiasis-causing flies (Diptera: Calliphoridae).

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The family Calliphoridae is formed by myiasis-causing flies, being known for its economic, medical, sanitary and forensic importance. Ecological similarities and the lack of morphological diagnostic traits may cause problems in the identification of species, especially for larvae. The mitochondrial DNA (mtDNA) is providing important clues for species identification and variability characterization. The control region (CR) represents the major noncoding region of the mitochondrial genome, with a bias towards A+T content. The CR has been divided in two different subregions, including the A domain with conserved sequence blocks and a hipervariable B domain. The entire CR could be recovered in two separated PCR reactions using a specific and universal primers combination. The efficient PCR amplification, sequencing and analysis of nine Calliphoridae species has shown a complete tRNA^{Ile} duplication into the B domain of four *Chrysomya* species: *C. albiceps*, *C. bezziana*, *C. chloropyga* and *C. megacephala*. The tRNA^{Ile} duplication could be used as a diagnostic marker for *Chrysomya* genus identification, since the duplication has not been reported for other related Calliphoridae genus. The characterization of the A domain for Calliphoridae species will allow the evaluation of this region as a molecular marker for evolutionary studies and for identification of Calliphoridae species.

Parasitoids of nuisance flies on Australian cattle feedlots

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Populations of adult and immature nuisance flies and associated parasitoids were monitored on three Australian feedlots with one southern Queensland (SQ) feedlot being continuously monitored over two years. In the SQ feedlot both sentinel pupae (cultured *Musca domestica* exposed for 1-2 weeks at various sites) and feedlot pupae, consisting largely of *M.domestica* (85%) and *Stomoxys calcitrans* (12%), were examined for parasitoids. The 4.2% of the sentinel pupae parasitised produced 7 species of parasitoids, with *Muscidifurax raptor* (52%) and *Spalangia cameroni* (39%), predominating. Parasitoids emerged from 10.3% and 11% of the *M.domestica* and *S.calcitrans* feedlot pupae respectively, with the emergence pattern indicating that both species were equally parasitised. Unlike the parasitised sentinel pupae, for feedlot pupae *Spalangia* spp. constituted 90% of the parasitoids with *S.endius*, *S.nigroaenea* and *S.cameroni* comprising 50%, 22% and 18% respectively. *M.raptor*, *Trichomalopsis* sp and Diapriids made up the remaining 10%. Some parasitoid preferences for various feedlot sites were evident but *S.endius* was the most common species found at all sites. Parasitoids, largely *S.endius* and *S.cameroni* were seen in 9% and 12% of pupae from feedlots in central NSW and central Qld respectively. Parasitoids appear to be one of the more important biological control agents on Australian feedlots.

Successional aspects of the sarcosaprophagous arthropods fauna associated to rabbit corpses in the nearing areas to Tenjo (Cundinamarca)

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The succession of adult sarcosaprophagous arthropods was studied using baited Schoenly traps and rabbits as model. The efficiency of the traditional methodology (Ptrad) against the use of baited Schoenly traps (PTC) was comparated. Modifications to the Schoenly trap were made in the material of the trap, a communication with the substrate, interchangeable collector bottles and the collector funnels. During the 42 days of sampling 11.847 individuals were collected included in 99 species (97 in PTC and 63 in Ptrad); this includes 43 families (43 in PTC, and 32 Ptrad) belonging to five troffic rolls: necrophagous, opportunists, parasites, prayers and accidental. Also significant differences in the species abundance, richness, diversity and dominance were found between PTC and PTrad. By multivaried analysis techniques five stages were determined: Fresh (72 hours), Active decomposition (78h), Advanced Decomposition (330h), dry Remains (288h) and Bones (144h). These stages deferred in duration with those found in other works as a result of the sampling efficiency. Significant differences between the five stages were found in the abundance of individuals, species richness, diversity, dominance and community composition. In conclusion the PTC showed to be more effective in collecting arthropod fauna and superior to Ptrad for evaluating the adult sarcosaprophagous succession process.

Altered colonization of dung from cattle treated with doramectin, eprinomectin, ivermectin or moxidectin

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Endectocides (e.g., abamectin, doramectin, eprinomectin, ivermectin, moxidectin) are used worldwide to control internal and external parasites affecting cattle. After application, most of the dose is excreted in the faeces of the treated animal where residues can either enhance or reduce the attractiveness of dung to coprophilous insects. By influencing the number of colonizing insects, residues may indirectly alter the rate of dung pat degradation. The current study compared collections of insects from pitfall traps baited with dung of untreated cattle versus dung of cattle treated 1 wk earlier with topical applications of ivermectin, doramectin, eprinomectin or moxidectin. The effect of residues on colonization previously has not been reported for the latter three compounds. In addition, these results are the first to compare directly the effects of faecal residues on colonization, when potential confounding factors of drug formulation, cattle diet, insect species, and time are held constant.

On the ubiquity and phylogeny of *Wolbachia* in lice

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Wolbachia are intracellular bacteria of interest, because they profoundly affect the reproductive fitness of their insect hosts. Here, we provide the first report of *Wolbachia* in the two insect Orders Anoplura (sucking lice) and Mallophaga (chewing lice). We document the apparent ubiquity and diversity of *Wolbachia* in these orders with infections detected in each of 25 tested populations of lice, representing 19 species from 15 genera spanning eight taxonomic families. Phylogenetic analyses indicate a high diversity of *Wolbachia* in lice, as evidenced by the identification of 39 unique strains. Some of these strains are apparently unique to lice, whereas others are similar to strains infecting other insect taxa. *Wolbachia* are transmitted from infected females to their offspring via egg cytoplasm. Hence, the evolutionary radiation of *Wolbachia* strains is predicted to correspond with their host taxa. This prediction is met with strains in the *Wolbachia* taxon Supergroup B, which form a monophyletic clade. In contrast, strains in Supergroup A exhibit phylogenetic discrepancies that previously have been attributed to horizontal transmission between host species, possibly via a parasitoid vector. We interpret such discrepancies in the current study as evidence of genetic recombination between *Wolbachia* strains or between *Wolbachia* and non-*Wolbachia* bacteria.

Host preference in *Anopheles gambiae*: decision between plant and human volatiles in a wind tunnel

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Both sexes of *An. gambiae* have small energy reserves at emergence, and at 27°C they begin to die within 2 days. Thus, 1 night after emergence is a critical time to obtain food. In a 2-choice wind-tunnel olfactometer, they oriented in significantly greater numbers toward honey, a source of nectar-related volatiles, than to human-foot volatiles. This was true both when the volatiles were paired with blanks and when they were presented simultaneously. In the latter case, the difference in response was greater, indicating direct competition between the two kinds of volatiles. After 5 days of sugar feeding and mating, males still strongly preferred honey volatiles, but females responded almost exclusively to human volatiles. These results suggest that under some conditions in the field, females take their first meal preferentially from plants. Females given sugar on the night of emergence exhibited stronger responses to human volatiles the following night, indicating that energy status itself promotes blood feeding, independent of maturation.

Phylogenetic relationships of midges of the *Culicoides obsoletus* species complex (Diptera: Ceratopogonidae) based on ITS2 rDNA sequences

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The midges of the genus *Culicoides* Latreille, 1809 include species that are vectors of orbiviral diseases of bluetongue and African horse sickness that cyclically decimate livestock in the Mediterranean Basin. Recent outbreaks of these diseases in regions where the known Afro-Asiatic vector, *C. imicola,* is absent has incriminated other *Culicoides* species belonging, amongst others, to the Obsoletus Complex of the subgenus *Avaritia* Fox, 1955. The lack of taxonomic clarity concerning these potential vectors stimulated the molecular analysis of 21 *Culicoides* population samples from 18 localities spread throughout mainland Italy. Nuclear ribosomal DNA ITS2 sequence variation was used to determine the genetic differentiation within and between the members of the Obsoletus Complex and related species, by means of AMOVA and phylogenetic analyses. Although morphological discrimination of the members of these complexes is extremely difficult, the molecular analysis clearly demonstrated a high degree of divergence between most of the species. Also evident is that the subgenus *Avaritia* comprises at least three species complexes, all of which are separate from the Imicola Complex which was represented by *C. imicola* as the outgroup. Furthermore, a simple PCR assay was developed which rapidly discriminates between the species of the complex, a prerequisite for vector identification.

"Differences in sensitivity to action of insect hormones analogues and agonists in several populations of *Culex pipiens* s.l. caused by insect pheromones"

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"Increasing interest for effective insect control, especially of hematophagous insects is provoked by number of diseases transmitted by some species from these insect groups. One of many ways how to control these species is the use of analogues and agonists of insect hormones. These analogues were tested against larvae of different strains of *Culex pipiens*. Larvae of *C. pipiens molestus* and *C. pipiens quinquefasciatus* originated from different countries were used in experiments. *Culex quinquefasciatus* is serious hygienic problem - vector of filariosis. Larvicidal activity of the methoprene, 20-hydroxyecdysone RH-5992 and RH-2484 were tested. All used compounds show different activity among experimental strains. In our laboratory strains - originated from Cuba, Iraq, India (two strains), Taiwan all tested compounds had dose depended activity. Toxic effect, morphological effects - inhibition of larval pupal or pupal-adult transformation and delayed pupation were observed. Very high activity of methoprene and 20-hydroxyecdysone were found in Cuban strain. On the other hand RH compounds were very active in Taiwanian strain. For example 20-hydroxyecdysone was still very toxic at 5 ppm concentration in Cuban strain, while in the Indian strain same toxic effect was caused at concentration 100 ppm. Similar results were obtained with methoprene RH-5992 and RH-2485.
"

Gene expression in ticks induced by blood meal and lyme borreliosis spirochetes

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The castor bean tick, *Ixodes ricinus,* is the principal European vector of *Borrelia burgdorferi* sensu lato, the causative agent of Lyme borreliosis (LB). Differentially expressed genes that underlie the host response of *I. ricinus* to *B. burgdorferi* have not been investigated prior to this study. The characterization of genes that are induced following an infected blood meal is essential for gaining an understanding of the molecular mechanisms that underlie vector-pathogen interactions. Subtractive hybridization was used to isolate tick genes that were induced in whole ticks following a blood meal and a blood meal on a *B. burgdorferi* infected guinea pig. Novel cDNA clones with similarity to cytochrome c oxidase, salivary secreted protein, actin, glutenin and a cysteine protease pro-peptide were induced following a blood meal. Novel cDNA clones with similarity to thioredoxin peroxidase(s), dolichyl-phosphate glucosyltransferase, glutathione-S-transferase, defensin, ML domain containing protein and von Willebrand factor were induced following *B. burgdorferi* infection. Northern analysis and RT-PCR were used to verify that these genes were differentially expressed in ticks and to determine the site of their expression. (Supported by the Grant Agency AS CR No. A6022306.)

Immune proteins in the soft tick *Ornithodoros moubata*

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Three proteins playing a role in the immune system and/or pathogen transmission were isolated from the soft tick *Ornithodoros moubata*.:1. An abundant tick plasma protein was isolated and identified as a protease inhibitor of a_2-macroglobulin (a_2M) family. Tick a_2M - **TAM** has an atypical structure in that it is composed of two noncovalently bound subunits of about 180 kDa. Each subunit is post-translationally processed in two disulfidically bound chains in a manner similar to the C3 and C4 components of complement. TAM sequence is closely related to the *Limulus* a_2M. TAM possesses at least 4 variants of bait region domain created likely by alternative splicing. 2. A sialic-acid binding lectin named **Dorin M,** apparently produced by tick hemocytes has been purified from tick plasma. Cloning and sequencing of the DorinM gene revealed that the protein shares significant similarity with the family of fibrinogen-related proteins (especially tachylectin 5A, B from the horseshoe crab and ficolins of vertebrates) which function as pattern-recognition molecules. 3. An anti-bacterial protein **lysozyme** was isolated from gut contents of engorged ticks. The enzyme is apparently upregulated upon bloodmeal. The features of lysozymes with anti-bacterial as well as digestive function seem to be combined in the tick gut lysozyme sequence.

Characterization of hemolipoproteins from ticks.

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Hemolymph proteins in ticks have not been completely studied. The hemolymph protein most studied in ticks has been vitellogenin (Vg). Besides Vg, another major hemolymph storage protein, a carrier protein (DvCP) and heme-binding lipoprotein (HeLp, for Heme LipoProtein) has recently been studied in two hard tick species but not in soft ticks. DvCP from *Dermacentor variabilis* is identical to a heme-lipoprotein (HeLp) from *Boophilus microplus*. An analogous protein to DvCP was reported in the soft tick, *Ornithodoros parkeri* (OpCP) but its tissue distribution and biochemical properties have not been fully understood. Further characterization of OpCP and biochemical characterization of major hemolipoproteins from other important tick species is in progress. Biochemical and molecular comparison of major hemolipoproteins across tick species and their significance to tick biology will be discussed.

Molecular detection of several tick-borne pathogens from ticks in Japan.

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Detection and analysis of several tick-borne pathogens, including *Rickettsia, Ehrlichia, Hemoplasma, Borrelia* and *Babesia,* from ticks recovered from dogs and cats in Japan were attempted using molecular techniques. A total of 1136 tick samples from dogs and 132 from cats were examined for tick-borne pathogens by using genus- and species-specific PCR, and sequence analysis. Some well-known pathogens such as *Anaplasma platys, Mycoplasma haemofelis, Mycoplasma haemominutum, Babesia gibsoni, Babesia canis* and *Borrelia garinii*, were detected and some new information on their distribution in Japan were added. DNA fragments of some new species were also found in the present study. Several rickettsial DNA closely related to *Rickettsia japonica, R. helvetica, R. canada,* or *R. akarii* were detected. Ehrlichial DNA closely related to a nevel *Ehrlichia* sp. detected in Tibet, Thailand and Africa were detected from *Hamaphysalis* ticks. Unique sequences that were similar to those of *Babesia odocoilei* and *Babesia divergens* were also obtained from *Ixodes ovatus*. *Borrelia valaisiana*-like DNA were detected from *Ixodes granulatus* in Okinawa. The combination of PCR and sequence analysis used in this study was demonstrated to be useful for detection of tick-borne pathogens from ticks. These can be excellent tools for epidemiological study of tick-borne diseases.

Species composition and dispersal of nuisance flies breeding on poultry farms in southern Australia

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"Ability to disperse and vectorial capacity makes flies a significant biosecurity and food safety risk on poultry farms. Although the species and biology of flies breeding on these farms has been widely studied overseas there have been few studies in Australia. We monitored numbers and species of flies breeding on egg farms in southern Australia over a 12 month period and assessed the dispersal of the major species using mass marking of resident fly populations with fluorescent dye. The little house fly, *Fannia cannicularis* and to lesser extent false stable fly *Muscina stabulans* were the major species for most of the year. In contrast to most overseas studies, *Musca domestica* was absent or present in only low numbers at most times. Most *F. cannicularis* remained within 700 m of the farm whereas marked *M. stabulans* were trapped at distances of up to 2 km. Modelling of total trap catches by distance predicted maximum dispersal distances of 1.6 km for *F. cannicularis* and 2.4 km for *M. stabulans.*

y-Season mosquito breeding associated with irrigation at Kununurra, Western Australia: potential impact on arboviral disease transmission.

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This study describes a preliminary investigation into the role of the Ord River Irrigation Area at Kununurra in mosquito production during the dry season. Specifically, the study aimed to test the hypothesis that irrigation has provided the opportunity for year-round breeding of arbovirus vector mosquitoes. A ten-day adult and larval mosquito sampling survey revealed that breeding was occurring during August, the iest month of the year at Kununurra, supporting the hypothesis that mosquitoes can breed year-round. Significantly larger numbers and percentages of adult *Culex annulirostris*, an important disease vector in Australia, were collected within the irrigation area (n=3706, 44.6%) compared to reference areas outside the irrigation area (n=363, 9.8%) (mean difference 76.2 per trap per night; 95% CI 38.6, 113.7; p<0.001). Larval *Cx. annulirostris* were also collected at several sites within the irrigation area, whereas none were collected at reference areas. Year-round mosquito breeding at Kununurra may increase human health risk from arboviral disease, as has been borne out by recent elevated notifications of Ross River virus disease in the late y-season compared to other towns in the Kimberley region. Mosquito management needs to be given a high priority to ensure this health risk is not further exacerbated. Strategies to reduce mosquito breeding should primarily focus on modification of breeding habitats and altering of irrigation protocols.

Comparative analysis of Calliphoridae complete mitochondrial genome sequences with other dipterans.

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In the view of the medical, veterinary, economic and forensic importance of Calliphoridae species, a comparative analysis of their nucleotide sequences with other dipterans would be useful for the molecular evolutionary studies of this family. Mitochondrial DNA (mtDNA) has been used mainly because of its maternal inheritance, accelerated rates of substitution, and lack of recombination. Of the 1020 Calliphoridae species described, 45 are listed in GenBank, including two complete mitochondrial genomes of the species *Cochliomyia hominivorax* and *Chrysomya chloropyga*. The comparison of the nucleotide content, skewness, aminoacid sequences, and codon usage between the mtDNA of Calliphoridae species and among the dipteran species can provide important features for studies in different taxonomic levels. Until now, the *cox1* gene is the most used molecular marker for studies among Calliphoridae and across a broad range of insects, but the comparison of individual and concatenated sequences of another mitochondrial genes among dipteran species have shown distinct degrees of conservation, indicating that some of them also should be employed as reliable molecular markers. The complete mitochondrial genomes are an useful source for improving primer designing, molecular identification of species, and for elucidate the potential of individual and combined genes for molecular systematic studies of Diptera.

Dispersal of house flies in the agricultural and urban environment: population dynamics Study using microsatellite markers

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The house fly, *Musca domestica* L., is a synanthropic species with worldwide distribution. Due to their unrestricted movement, mode of feeding, developmental habitats, and attraction to places where food is prepared and stored, house flies greatly amplify the risk of human exposure to food-borne and other pathogens. Flies that develop in animal productions where antibiotics are frequently used for prophylaxis and growth promotion carry antibiotic-resistant bacteria that maybe transmitted to food intended for human consumption, leading to illnesses that are difficult to treat. However, no studies have been undertaken to quantify the extent of house fly movement between rural to urban areas. Therefore, our objective is to examine the population genetics of house flies and estimate the extent of dispersal/migration from rural to urban areas and *vice versa* in Kansas. As a first step, we developed polymorphic microsatellite loci for house flies. Here we report on the isolation, characterization, and optimization of microsatellite loci and preliminary results on population dynamics of house flies.

Comparative study on the nocturnal behavior of *Aedes* mosquitoes using a new device equipped with photoelectric sensor

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The nocturnal behavior of non-blood fed *Aedes aegypti* and *Ae. albopictus* females was observed using a new automatic recording device equipped with photoelectric sensor. Carbon dioxide, heating and the contrast of black and white color were used for the attractive factors for mosquitoes. The nocturnal host seeking activity positively correlated to the increasing light intensity in both species. *Ae. aegypti* was found to be more sensitive to light than *Ae. albopictus.* The threshold of light intensity for activation of the nocturnal host seeking activity was < 0.1 lux (ca 0.01 foot candle) in *Ae. aegypti* and > 10 lux (ca 1 foot candle) in *Ae. albopictus,* respectively. Complete darkness in the daytime, on the other hand, deactivated the host seeking activity of *Ae. aegypti,* irrespective of their intrinsic circadian rhythm, which might well explain the importance of the sense of sight in *Ae. aegypti.*

Rainfall events determine the abundance of carrion feeding beetles in a West African savanna

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Using carrion beetles on goat and warthog skins as a model system, we studied the influence of single rainfall events on the abundance and flight activity of insects in a tropical savanna (Parc National de la Comoé, Côte d'Ivoire). Although the attraction of the skins declined with time (due to degradation and desiccation), the freshness of the resource was not the major factor determining the abundance of carrion beetles. After rainfall events the number of beetles sharply increased, even with older skins. We recorded the same effect in regularly watered skins (simulating continuing freshness), showing that the humidity of the resource was not responsible for the peaks. The sharp abundance peaks after rainfall events is of relevance for both ecological field studies which have often a standardized sampling programme ignoring weather, and for forensic studies relying on a predictable succession of carrion users which can be strongly influenced by weather.

Environmental parameters of habitat on breeding behavior of *Aedes albopictus* in Chonju, Korea

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Aedes albopictus during present studies has been associated with tree, automobile tires, very rarely apartment basements and open outdoor garbage containers. However, a contrast to the habitats of this insect is found; at one stage it seems that this mosquito can live in almost all places wherever there is debris, standing water regardless of the pH acidic or alkaline. Whereas, in certain cases it is evident that this insect is confined to a particular habitat, during the present investigation, it was quite evident that the outbreak of adult *Aedes* is very much dependent upon the percent humidity and temperature. The artificial habitats, that is abandoned tanks, cans, stray automobile tires if they are close to plantation or are in shade for most of the photoperiod they tend to be positive breeding sites for *Aedes* sp. In other situations, the same habitat in open with sunlight throughout the day tends to be negative breeding site. Water level in containers is also known to effect the population and species of the mosquitoes. Temperature and precipitation have been found to impose serious effects not only on the distribution of *Aedes* but also the size of the immature and adults as well.

Sheep lice control with entomopathogenic fungi

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Preliminary investigations into the potential for the biological control of the sheep body louse (*Bovicola ovis)* by the entomopathogenic fungi *Metarhizium anisopliae* and *Beauveria bassiana* are reported. Scanning electron microscopy was used to investigate the uptake of inoculum from the wool by lice and bioassay methodologies using a wool substrate were developed to test pathogenicity. Fungal strains were identified that caused a 100 % mortality of lice after 5days. The level of lanolin in the wool and stage of louse development had a significant effect on mortality rates but there was no effect of temperature within the range of 31^{o}C to 38^{o}C. Further studies are investigating factors that influence the pathogenesis of fungal disease in lice.

The mitochondrial DNA control region of Muscidae species (Diptera: Calyptratae): evolution and structural organisation

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The control region of insect mitochondrial DNA (mtDNA), also called A+T-rich region, is an interesting target for studies regarding the structure and evolution of the mtDNA. This non-coding region has an biased AT content, putative regulatory elements for transcription and replication processes, high levels of nucleotide substitutions and ins/dels events, and structural instabilities related to the organisation of internal sequence elements and its flanking genes. The sequence of *Haematobia irritans* and *Musca domestica* (Diptera: Muscidae) control regions have been determined and the sequencing of *Stomoxys calcitrans* control region is being conducted. Conserved sequence elements described for insect mtDNA control regions (such as poly-T streches, TA(A)n in tandem repeats, 'stem-and-loop' secondary structure and G+C-rich region) and conserved sequence blocks previously described for other Calyptratae species (myiasis-causing flies) have been identified in the Muscidae mtDNA control region. The share of such structural homologous elements among Muscidae and myiasis-causing flies suggests that Muscoidea and Oestroidea superfamilies have a similar regulatory mode for mtDNA transcription and replication processes. Intraspecific analysis of *H.irritans* populations using control region as a molecular marker are being conducted and length variation among specimens from Brazil, Argentina, Uruguay, Venezuela and Malaysia were already identified.

Key diagnostic characteristics of the third instar larvae of *Calliphora croceipalpis* and *Sarcophaga cruentata*

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Following the death of a human, various insects involved in the decomposition process invade the corpse. This invasion takes place in an orderly sequence, called insect succession. This poster deals with those morphological characteristics used to distinguish between third instar larvae of the secondary invader *Calliphora croceipalpus* (Jaennicke) and the tertiary invader *Sarcophaga cruentata* (Meigen). All aspects of larval morphology were studied, but specific note was taken of the following aspects for diagnostic purposes: nature of the spines and spine band, anterior spiracles, posterior spiracles and the spiracular atrium of the posterior spiracles. The distinguishing characteristics of *S. cruentata* were the deeply recessed atrium cavity and the scalloping of the inner margin of the posterior spiracular plate. In *C. croceipalpus* the distinguishing characteristics were the shallow posterior spiracular atrium and the orientation of the spiracles. These key characteristics were compared to the key characteristics of the third instar larvae of the other species that form part of this study *viz. Lucilia cuprina* (Wiedemann), *Chrysomya chloropyga* (Wiedemann), *Chrysomya marginalis* (Wiedemann) and *Chrysomya albiceps* (Wiedemann).

The effect of temperatue on the development of the immature stages of *Hemilucilia segmentaria* (Fabricius) and *Hemilucilia semidiaphana* (Rondani) (Diptera: Calliphoridae).

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Diptera larvae are among the most important carrion insects, because they are one of the most frequent and abundant organisms involved in animal decomposition process. This work objectived to determine the effect of temperature on the development of immatures of *Hemilucilia segmentaria* (Fabricius) and *Hemilucilia semidiaphana* (Rondani), two species of Neotropical blowflies commonly associated with animal carrion in rural and natural areas of Brazil, being two of the most important forensic indicators in the neotropics. The rearing of the immatures of both species was done using growth chambers with temperatures set at 10, 15, 20, 25, 30 and 35ºC, 12h photoperiod and 60 ±10% relative humidity. The mean developmental time from egg to pupation decreased as temperature increased. There were also differences in larval length, width and weight. These data were used to determine logistic growth curves to be used in estimating the age of the immatures of both species.

Larval competition differentially affects arbovirus infection and dissemination in *Aedes albopictus* **and** *Aedes aegypti*

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We determined the species-specific effects of intra- and interspecific larval competition between the mosquitoes *Aedes albopictus* and *A. aegypti* on adult susceptibility to infection with the arbovirus Sindbis. We found higher infection rates and whole-body viral titer, but lower dissemination rates, for *A. albopictus* than for *A. aegypti*. For *A. albopictus* but not *A. aegypti*, intra- and interspecific competition resulted in greater infection rates compared to low-competition rearing conditions. Size-dependent and size-independent effects of competition influenced arboviral infection and dissemination of *A. albopictus* in opposite ways. At a single level of competition (i.e., at a single density), higher viral titers occurred in larger *A. albopictus,* but between competitive treatments adults from low-competition larval rearing conditions had greater mean body size and lower body titer and a lower proportion infected, compared to adults emerging from high-competition conditions. Competition experienced by larvae had no detectable effect on acquisition of infection by *A. aegypti*. Higher dissemination rates and less sensitivity of *A. aegypti* females to effects of larval competition are consistent with the greater role of this species as a vector of viruses to humans.

Mitochondrial DNA variability in screwworm fly from Uruguay.

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The New World Screw-worm fly (NWS), *Cochliomyia hominivorax* (Diptera: Calliphoridae), is an obligate ectoparasite that causes primary myiasis in warm-blooded vertebrates throughout Neotropical regions. In Uruguay this pest is responsible for important economic looses to cattle breeding sector. In this work RFLP and PCR-RFLP analysis of mitochondrial DNA (mtDNA) were used to examine the genetic variability and population structure of six NWS geographic populations from Uruguay. RFLP analysis was performed using 5 restriction endonucleases and there were found 12 haplotypes. PCR-RFLP was used to survey mtDNA variation in control region and Cytochrome Oxidase subunits I and II. There were found 8 haplotypes utilizing 3 restriction endonucleases. The two techniques revealed the same pattern of haplotypes distribution, three haplotypes widely distributed and the remaining locally distributed. DNA diversity and divergence were estimated and the preliminary results revealed high genetic variability but no population subdivision. Our data are being compared with previous works that analyzed Libya samples collected during 1988 infestation in order to elucidate the origin of the introduction. The characterization of NWS intra specific genetic variability at the geographic level is necessary to monitoring the expansion in the current distribution and into screw-worm free areas. Financial support: FAPESP, CNPq, IAEA.

Paternal genome elimination in the human body louse, *Pediculus humanus* Linnaeus, 1758.

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Eight F_1 and two F_2 families of human body lice, *Pediculus humanus*, were bred to examine the inheritance of alleles in this species. Alleles at four autosomal microsatellite loci segregated in two ways in these lice: (i) in a Mendelian fashion i.e. diplo-diploidy, and (ii) in a non-Mendelian fashion. We interpret the latter as paternal genome elimination, where parental males transmit, exclusively, their maternal genome to their offspring (functional haplodipoidy). Further investigations of this extraordinary finding may provide insight into how haplodiploidy evolves in insects.

Anthropogenic ecological change and insects of health concern in New Zealand.

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Key components of anthropogenic ecological change include land conversion to human uses and the fragmentation and disturbance of native ecosystems. This is combined with the spread of alien species and human commensals and loss of locally evolved species. New Zealand is a microcosm of some global issues in this regard. Our research examines ecological change and changes in the habitat, distribution and productivity of native and exotic mosquitoes, the relation of mosquitoes and potential arbovirus vector species, and the disease burden of bee & wasp stings in New Zealand.

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CJ McMeniman, SC Barker
Department of Microbiology and Parasitology, and Institute for Molecular Bioscience, University of Queensland, St Lucia, QLD 4072, Australia; c.mcmeniman@uq.edu.au

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An *in vitro* larval immersion microassay for screening candidate Acaricides

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Acaricide resistance in *Boophilus microplus* is rising dramatically in regions of the world where this ectoparasite is prevalent, posing significant economic burdens on cattle production and productivity in Central and South America, South Africa and Australia. While *Boophilus* has been eradicated from the United States, acaricide resistance will increase the threat of reintroduction into southern portions of the U.S. New chemical agents are needed that combine potent activity with novel modes-of-action not exhibiting cross-resistance to existing acaricide classes. Currently, none of the available *in vitro* tick bioassays offers the flexibility, speed, and sensitivity required to rapidly screen large numbers of compounds for candidate acaricides. We have optimized and characterized a larval immersion microassay (LIM) that offers superior sensitivity, the flexibility to accommodate multiple solvents and detergents, and a robust capability for screening compounds in a relatively short period of time using a minimal quantity of test article for evaluation. Dose-mortality studies were conducted against multiple ixodid ticks, including ug-resistant *B. microplus*, with representative formamidine, organophosphate, synthetic pyrethroid, pyrazole, macrocyclic lactone and carbamate acaricides. The rationale for using this method to screen chemical or natural product libraries and identify, characterize, and prioritize compounds for further development will be highlighted.

Incidence of bed bugs (Hemiptera: Cimicidae) in shared Accommodation In Sydney, Australia

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Bed bugs have remerged as significant pests in Australia and worldwide in the past ten years, The results of a survey of the incidence of bed bugs in shared accommodation in Sydney is presented. The survey examined the incidence of the bed bugs and the relationship of the infestation to a number of factors including, season, building construction and furnishings. It also examined control methods, their frequency and effectiveness. The results demonstrated widespread incidence of bed bugs with the majority of premises reporting an infestation in the previous 12 months and with many premises having multiple infestations throughout that period. Current control measures did not seem to be eliminating the problem as recurrence of the pest was common. A more comprehensive control strategy for bed bugs needs to be developed to manage this pest in shared accommodation.

Characterization of the enzymatic activities present in extracts of various mites.

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Many of the previously-characterized allergens of house dust mites are known to be proteases and this enzymatic activity is thought to contribute to their allergenicity. Other astigmatid mites are also known to be allergenic but little or nothing is known about their enzymatic activities. The purpose of this study was to characterize the enzymatic activities present in extracts from 9 species of astigmatid mites, several of which have not previously been investigated. Extracts were prepared from 5 stored product mites (*Chortoglyphus arcuatus, Lepidoglyphus destructor, Blomia tropicalis, Tyrophagus putrescentiae, Acarus siro*), 3 house dust mites (*Dermatophagoides farinae, D. pteronyssinus, Euroglyphus maynei*) and one parasitic mite (*Sarcoptes scabiei*). ApiZym strips were used to screen for the presence of 19 individual enzyme activities. Digestion of 9 other substrates was also evaluated by spectrophotometric or electrophoretic methods. All mites exhibited some form of phosphatase, esterase, aminopeptidase and glycosidase activity although their substrate specificities varied considerably. Scabies mites did not possess detectable serine peptidase activity nor were they able to hydrolyze gelatin or casein while all other mites exhibited these activities. Storage mites possessed enzymes capable of degrading the widest range of substrates while scabies mites had the most limited proteolytic capacity.

Klinotactic orientation of the northern fowl mite to heat

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The northern fowl mite (NFM) (*Ornithonyssus sylviarum*) is a cosmopolitan bird ectoparasite and a serious pest of the U.S. poultry industry. Mite infestations spread within a flock, even when direct bird-to-bird contact is prevented, and heat is a suspected orientation cue. NFM movement patterns were evaluated in a spatial heat gradient, as well as in an environment with temporal fluctuation of temperature (spatially uniform). Mites were filmed and the movement tracks were digitally analyzed for changes in velocity, angular shift and mean bearing. NFM significantly increased linear velocity when exposed to heat (35 °C) relative to control mites in ambient conditions (~27 °C). Mean track bearings were significantly associated with the position of the heat source in a spatial gradient, while track bearings of mites in ambient conditions were randomly directed. Temporal shifts in temperature from heated to ambient condition caused NFM to execute turns ≥ 90° within 5 seconds. Shifts in temperature from ambient to heated condition did not consistently cause NFM to execute turns ≥ 90°. These experiments indicate NFM orient to a heat source by comparing temperatures at different points in time (longitudinal klinotaxis) and executing turns based on internally stored (idiothetic) proprioceptive information.

Screening medicinal plant extracts for effects on *Aedes aegypti* (Diptera : Culicidae) and toxicity to a non target organism

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A preliminary study was conducted to find out the effects of the extracts of 112 medicinal plant species, collected from the southern part of Thailand, on *Aedes aegypti*. Studies on larvicidal properties of plant extracts against the third and fourth instar larvae of *Ae. aegypti* showed fourteen species with high toxicity. Six plant species were comparatively more effective against the larvae at very low concentration. These extracts demonstrated no or very low toxicity to guppy fish (*Poecilia reticulata*). Three plants with promising larvicidal activity, *Mammea siamensis*, *Anethum graveolens* and *Annona muricata,* were used to study of the impact of the extracts on the life cycle of *Ae. aegypti*. The results showed that these plants affected the reproductive potential of surviving adults by reducing the number of eggs laid and egg hatchability. The first instar larvae were very susceptible to *A. muricata* and the second instar larvae were susceptible to *A. graveolens* while the third and fourth instar larvae were susceptible to *M. siamensis*. These extracts delayed larval development and inhibited adult emergence.

The prevalence rate of *Leishmania* infection among *Phlebotomus* collected from Abardejh of Varamin Iran

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Cutaneous Leishmaniasis is a prevalent tropical parasitic disease, which is caused by Leishmania protozoa. The infection can be limited in immun-compitant person, but it could progress to a chronic and ulcerative disease in immuno-comprmised person. The reservoirs are carnivore and rodents and the vectors are Phlebotomus. Abardejh of Varamin is located in southeast of Tehran and there is a tropical ecosystem in its eastern border. There is a suitable condition by the Tamarisk and rodents exist for *Phlebotomus* activity. In this study we investigated the prevalence of Phlebotomus population in Abardejh district. The collected sandflies by funnel trap from rodents' barrows transferred to the department of Parasitology at Pasteur Institute of Iran .The sampling was continued during the spring and summer in periodicals dates. The collected sand flies were studied to define the genius and species before parasitological culture in NNN media. The data revealed that the maximum activities of Phlebotomus were in Jun and July months. The results indicated the high prevalence rate of infection among the sand flies. This could be a matter of debate because of human and agricultural environments are located closely to this district. Therefore application of chemical control, environmental sanitation seems to be required to control of tranfection from sand flies to man.

The molecular identification of closely related calliphorids (Diptera: Calliphoridae) of forensic importance.

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Blowfly larvae (Diptera: Calliphoridae) are one of the most useful groups of insects utilised during the investigation of human deaths because they are among the first colonisers of a cadaver. The identification of such larvae is a critical step when estimating the the postmortem interval (PMI), as the rate of larval development can vary substantially between species. Two blowflies that have the potential to feature in forensic investigations are *Chrysomya semimetallica* and *Ch. latifrons.* The identification of these species is hampered by their almost indistinguishable morphologies, even as adults. The species were differentiated on the basis of two highly variable regions of the ribosomal DNA genes – the internal transcribed spacer (ITS) sequences. The amplification of ITS2 from both species was readily accomplished, however only ITS1 from *Ch. semimetallica* amplified successfully. The ITS2 differences correspond to 1.22 % sequence divergence between the species. A rapid, cost-effective strategy for species identification would involve PCR amplification of the ITS regions, followed by restriction fragment length polymorphism (RFLP) analysis. Despite the differences within ITS2, no restriction sites were observed that could differentiate the two species. Assessment of this possibility in ITS1 will be achievable once the remaining data is obtained from *Ch. Latifrons*.

A global presence of *Wolbachia* in human lice?

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Wolbachia are intracellular bacteria of general interest. The profound effects on the reproductive fitness of their insect hosts have resulted in plethora of phenotypes, which still await a full characterisation. Infections of *Wolbachia* in lice affecting humans are of particular interest. Endosymbionts of lice have a great potential for the control of these insects. Previous research in Canada suggests that infections of *Wolbachia* occur in all species of lice (Anoplura, Mallophaga). Independent and concurrent research in Wales provides substantiating evidence. The current study represents the subsequent joint efforts to test for the ubiquity of *Wolbachia* infections in populations of human lice (i.e, head lice - *Pediculus humanus capitis*, body lice - *Pediculus humanus humanus*, pubic lice - *Pthirus pubis*) from different continents.

Field efficacy of a personal insect repellent based on essential oils from Australian native plants against the mosquito (Diptera: Culicidae) and the biting midge (Diptera: Ceratopogonidae)

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Field trials were carried out in a wet tropical climate at separate sites in Cairns, Far North Queensland, Australia in Autumn 2002. One site consisted of a fresh water creek infested by mosquitoes and the other site a mangrove wetland infested by biting midges. Both studies were carried out during daylight hours and were conducted by periodically exposing human volunteers with bare lower legs (knee to ankle) to mosquitoes or biting midges over a four-hour period. Counts were made of mosquito landings over a five-minute period and biting midge landings over a three-minute period. Both legs of each volunteer were used to give five replicates of the repellent formulation and five replicates of the untreated control. Against mosquitoes, primarily *Verrallina carmenti* and biting midges, primarily *Culicoides ornatus*, the formulation significantly reduced the landing rate compared to untreated legs over the entire four-hour study period. This study demonstrated that this formulation provided protection against mosquitoes and biting midges for up to four hours after application.

Natural promastigote infection of *Phlebotomus caucasicus* in a new focus of visceral leishmaniasis in north west of Iran

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An investigation was carried out in Ahar County (a new focus of Visceral Lieshmaniasis) in order to determine the vector (s) of disease. This study was done in eastern Azarbaijan, North-west of Iran during the summer of 2003. Sand flies were collected using sticky papers, CDC light traps as well as aspirators. All of the collected sand flies were dissected in a op of normal saline for surveying of promastigote. Head and the two last abdomen segments of specimens were mounted in a op of Puri,s medium for species identification. Totally, 215 sandflies were collected including: *Phlebotomus kandelakii* (57.2%), *P.perfiliewi* (18.1%),*P.caucasicus* (11.2%), *P.sergenti* (7.9%), *P.papatasi* (3.3%) and *P(.adlerius)sp.*(2.3%). Only 1 out of 24 *P.caucasicus*(4.2%) was found naturally infected with parasites. This is the first infection report of *P.caucasicus* in this region. The ecology and some behavioural aspects of the vector will be discussed in details.

Application of a new *Aedes* surveillance method in Botucatu, Brazil.

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In Brazil, infection by dengue virus has increased significantly in the last two decades, after the reintroduction of mosquito vector *Aedes aegypti*. Epidemiological surveillance remains the only way to prevent dengue outbreaks. Currently, the control program applied in *Aedes albopictus* and *Aedes aegypti* populations are larval surveys. An interesting alternative is the using of a new control strategy named PCI (Premise Condition Index). This model relates the property condition, like house condition, yard condition, and degree of shade; to the occurrence of *Aedes sp* oviposition. The present study is based on the application of PCI in properties located in the urban area of Botucatu. Ovitramps have been set in 105 spots all over the city and their properties qualified. Results from properties analysis by PCI showed that 64.7% ones with scores 8 and 9 (considered bad condition houses) were positive to *Aedes albopictus* while only 19% of properties with scores 3 and 4 (good condition houses) were positive. This analysis demonstrated the accuracy of PCI method, since major occurrence of *Aedes sp* has been observed in properties with highest scores. There is a positive correlation ($r=0.76$, $p<0.025$) between PCI and positive house percentage, showing the significance of the method.

Molecular population genetics of *Aedes aegypti* in Brazil using RAPD

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Random amplified polymorphic DNA (RAPD) analysis technique has been used in studies of arthropod molecular taxonomy and in epidemiological studies of relatedness of geographic populations and vector movement. In Brazil, little is known about genetic structure among *Aedes aegypti* populations. We use RAPD to examine patterns of *Aedes aegypti* genetic structure in 11 populations from 6 Brazilian states. We found high genetic polymorphism (H_S = 0.38) and high levels of genetic differentiation exist both between population from different states (G_{ST} = 0.43) and between populations from cities in same state (G_{ST} = 0.41). These results indicate significant differentiation between *Aedes aegypti* populations in Brazil. There is a clear correlation between genetic structure and geographic localization showing that, if new populations are been introduced, they are not significant. Principal Component Analysis (PCA) of receive allele frequencies could separated populations in 3 different groups regarding geographic origin.

Biological characteristics of three domiciliated vectors of Chagas disease in Mexico.

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Mexico has a great population of triatomine bugs actually are reported seven genera and 24 species of Triatoma genus. Due to this great amount and diversity, surveys were made for to know ecotopes and biology of three species of the Triatoma genus. The objective is to show laboratory and fields observations made in regions where Trypanosoma cruzi vectors predominate: Triatoma barberi recollected in Queretaro state, T. pallidipennis in Morelos and T. dimidiata in Veracruz. T. barberi is an exclusive species of Mexico, it prefers walls, T. dimidiata is found in floor and the middle of the wall downwards, both in beooms, and T. pallidipennis prefers floor and clothes, also wildlife. Life cycle is 523 and 510 for Triatoma barberi and T. dimidiata, respectively and for to T. pallidipennis is 171 days. T. barberi feeds and defecates at the same time, the others two species defecated among 10 and 30 minutes. Infection index percentage was is 56.6 for T. barberi, 29.0 for T. pallidipennis and 15.7 for T. dimidiata. Percentage of infective forms in bugs feces were 76, 26 and 15 for T. barberi, T. pallidipennis and T. dimidiata respectively. T. barberi is the best vector of T. cruzi in Mexico

Droneflies occurring from effluent of manure in Hokkaido, Japan (Diptera: Syrphidae)

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Three genera and 5 species of droneflies (Diptera: Syrphidae), namely Eristalis (Eoseristalis) cerealis, Eristalis (Eoseristalis) roccica, Eristalis (Eristalis) tenax, Eristalinus (Lathyrophthalmus) tarsalis and Helophilus (Helophilus) virgatus, occurred from the ovitraps using the effluent of manure as attractant and media in Hokkaido, Japan.These droneflies were reared in the laboratory and the specimens of each developing stages were examined their morphological characters by the binocular and scanning electron microscopes. The following differences between the species were observed:Units of eggshell of H. virgatus were glabrous and those of the other 4 species were rhizoids.Sensilla of the 3rd instar larvae of E. rossica and E. tarsalis were spinous and those of the other 3 species were setae.Only E. tarsalis have spine rows at the 7th proleg and only H. virgatus have 3 pairs of lappets at anal segment of the 3rd instar larva.Trachae of the 3rd instar larva of H. virgatus are waved and those of the other 4 species were strait.Anterior spiracles of puparium of those species are all characteristic shaped.According to the combination of morphological characters above mentioned, it is easily distinguish the species at all developing stages.

Bioconversion with *Hermetia* to solve problems of manure accumulations and produce valuable feedstuff

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Black soldier fly, *Hermetia illucens* can eliminate many of the problems associated with large manure accumulations and with other putrescent wastes. The larvae consume fresh waste rapidly in managed systems and assimilate nutrients that otherwise might produce odors, pest flies and cause environmental pollution. Mature prepupae are self-collected for use as an animal feedstuff with about 42% protein and 35% fat. As a ration component, prepupae meal supported better fish growth than otherwise similar rations using fish meal. Data will be presented on this and other feeding studies with prepupae meal, amino acid and fatty acid profiles, elimination or reduction of several malodorous gasses, control of pest flies and reduction of waste dry matter by 50% or more and reduction in concentration of potentially polluting nutrients within that residue.

Prevalence of dengue virus in *Aedes* mosquitoes during y season by semi-nested reverse transcriptase-polymerase chain reaction (Semi-nested RT-PCR)

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Dengue hemorrhagic fever remains a major health concern in Thailand. Much effort has focused on the prevention and control of the disease. Detection of dengue virus infection rate in mosquito would evaluate dengue control programs and predict the epidemics of dengue hemorrhagic fever. To determine dengue virus infection rate in mosquito by Semi-nested RT-PCR. A total of 400 mosquitoes were collected from Rom Kao Community representing a crowded community and another 9 non-crowded communities in Bangkok. Mosquitoes were then divided into 40 pools, each contained 10 mosquitoes. A total of 391 *Aedes aegypti* and 9 *Aedes albopictus* were screened for dengue virus. Mosquito infection rate in Rom Klao community is 2.5% of mosquito pool equal to that found in non-crowded communities. Both groups are found to be dengue virus serotype 3. Our study suggests a circulation of dengue virus serotype 3 in both crowded and non-crowed communities, the infection rates of which are indifferent during dry season.

Field evaluation of bifenthrin 10WP and bifenthrin 80g/L SC on dengue vectors at high-rise flats in Kuala Lumpur , Malaysia.

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Bifenthrin 80g/L SC and Bifenthrin 10WP were evaluated against the sentinel sugar fed adults and 4th instar larvae of *Aedes aegypti* at 17 storey high-rise flats in Kuala Lumpur.The impact of both pyrethroid formulations on field populations of *Aedes aegypti* and *Aedes albopictus* larvae was monitored weekly using bottle containers.Both formulations of Bifenthrin showed adulticidal effects causing between 96% and 98% of *Aedes aegypti* mortality in the field compared to 18-19% in the control. However,both Bifenthrin formulations had not shown larvicidal effects when sprayed with ULV. This field trial indicated that both Bifenthrin formulations had potential for use in future dengue vector control programs.

Geographical range predictions of *Anopheles* mosquitoes in northern Australia using a genetic algorithm and data mining methods

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An appreciation of the environmental factors which influence the geographical ranges of mosquito vectors of malaria is of considerable interest from both ecological and epidemiological viewpoints. GARP (the Genetic Algorithm for Rule-Set Prediction) was used for ecological niche modelling of *Anopheles farauti* ss, *Anopheles farauti* 2 and *Anopheles farauti* 3 in northern Australia. Models were developed using 41 environmental layers together with species locality datasets derived from systematic mosquito surveys in northern Queensland and the Northern Territory. For each species the predicted ranges included almost all presence records and excluded areas in which that species was not found in repeated collections. The datasets were analysed with CART (Classification and Regression Tree Analysis) and KnowledgeSeeker to investigate associations between environmental variables and the record/no-record collection sites for each species. The outputs were used for further model development with GARP to produce accurate range maps of *A. farauti ss* and *A. farauti* 3 using only 6 or 7 of the most significant environmental layers identified by decision-tree software. Further data mining analyses showed that the use of simulated absence data in lieu of no-record collection data may highlight significant environmental variables in datasets which do not have records for species absence.

A new pediculicide based on isopropyl myristate

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New pediculicides need to be effective, safe, and to act differently from other products so as to prevent or delay resistance. Isopropyl myristate (IPM) is approved for many cosmetic uses and is thus safe for topical use. IPM is not known to be neurotoxic to insects, unlike most other pediculicides. IPM in cyclomethicone was tested against body lice and their eggs (*Pediculus h. humanus*) as surrogates for head lice (*P. h. capitis*) by the ASTM methods. IPM gave 100% knockdown at +1 hour and 88.8% at +4 hours. 24-hour mortalities were: 15% IPM; 68.8%, 50% IPM; 97.1%, 100% IPM; 100%. Ovicidal action was slight, with mortalities under 20% The mode of action is unresolved, but is likely to be related to IPM's spreading and penetrating action. Symptoms were, however, similar to those from a neurotoxic synergized-pyrethrin pediculicide; Rid®. Toxicity was lower in lice exposed orally to IPM (56.0%) than by direct contact with a treated substrate (90.4%). Direct contact gave signs of desiccation, oral exposure did not. A cuticular route of action is indicated. An IND has been submitted to FDA and Phase II clinical trials start in April 2004.

Development of new microsatellite markers in the New World screw-worm *Cochliomyia hominivorax* (Diptera: Calliphoridae)

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Microsatellites are a highly informative class of DNA markers, consisting of tandem repeats of 2-6 bp distributed throughout eukaryotic genomes. Microsatellite loci stand out as codominant and multiallelic markers with high polymorphism and expected heterozygosity value. Due to these characteristics, they were the molecular markers chosen to continue our efforts to investigate the genetic variability and population structure of the New World screw-worm (NWS) *Cochliomyia hominivorax* (Coquerel). This species is one of the most important parasitic insect pests in Neotropics, causing myiasis and, consequently, significant losses to livestock industries in these regions. In order to generate new microsatellite markers for NWS, an enriched genomic library for microsatellite inserts was constructed. Of 56 clones inserts sequenced, 30 had a microsatellite and of these, 10 sequences with repeats of di-, tri-, tetra- and hexanucleotides were selected. Primer pairs were generated for the ten selected loci and eight of them were successfully amplified in NWS DNA samples. Polymorphism of each locus will be evaluated and the informative markers, in combination with ten polymorphic microsatellite markers previously described, should provide high resolution for assessing the fine-scale genetic structure in NWS.

Genetic structure and diversity in new world screw-worm *Cochliomyia hominivorax* (Diptera: Calliphoridae) populations from Uruguay revealed by microsatellite markers

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The New World screw-worm (NWS), *Cochliomyia hominivorax* (Coquerel), is one of the most important parasitic insect pests that cause invasive myiasis in warm-blooded vertebrates. Because of the substantial economic losses to livestock rearing caused by this pest throughout the neotropics, an international effort has been involved in a program to eradicate the NWS from endemic areas of Central and South America and to prevent invasions into screw-worm-free areas. It is crucial to characterize the genetic variability of target populations in order to maximize the effectiveness of the eradication program. Ten polymorphic microsatellite loci previously isolated from an enriched library constructed for *C. hominivorax* are currently being used to investigate the genetic variability and population structure across its current geographic distribution. Preliminary results obtained from Uruguayan populations indicated a considerably high genetic variability. Statistical analysis indicated that most of the genetic variation is contained within populations, while low levels of differentiation were found between them. The investigation into the genetic variability and population structure of NWS from Central and South American populations should provide fundamental information for future eradication strategies.

Isolation of microsatellite loci in the blowfly *Chrysomya chloropyga* (Diptera: Calliphoridae)

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The species *Chrysomya chloropyga* (= *Chrysomya putoria*) (Wiedmann) is a synanthropic blowfly native from African regions. The adults feed and breed in carrion, feces, urban garbage and carcasses. Due to these biological features, *C. chloropyga* have great medical-veterinary importance acting as a vector of pathogens and is considered of potential forensic importance. This fly was probably introduced into Brazil around 1975 with the influx of Angolan refugees. Since then it has spread rapidly in Brazil and other countries of South America affecting the ecological scenario of the native Calliphoridae fauna. Genetic studies based on mitochondrial DNA revealed insufficient polymorphism at a fine scale. Since microsatellites are one of the most powerful markers for studying inter- and intrapopulation relationships, our goal is to develop a set of polymorphic microsatellite markers for *C. chloropyga*. A genomic library enriched for the motifs poli-AC/TG and poli-AG/TC was constructed. We are currently screening this library for microsatellite-containing clones. Of 54 clones inserts already sequenced, 26 had a microsatellite and 12 are useful sequences for designing primers. After polymorphism evaluation, these loci should contribute to population dynamics studies and for understanding the evolutionary context of the introduction of this species in the New World.

Survey of new world screw-worm *Cochliomyia hominivorax* (Diptera: Calliphoridae) for the presence of a point mutation associated with esterase-mediated organophosphate resistance

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The *Cochliomyia hominivorax* (Coquerel) is the main agent of primary myiasis in the South America, generating severe economic losses to livestock industry. In Brazil, its control has been done by the application of insecticides, but the inappropriate use of these chemical agents can select resistant flies. It has been shown in another species of the Calliphoridae family, *Lucilia cuprina*, that a Gly137®Asp substitution in the enzyme carboxylesterase (E3) converts its typical activity to an organophosphate (OP) hydrolase activity resulting in the development of OP resistance in this species. Preliminary efforts to isolate and characterize the E3 gene in *C. hominivorax* using primers previously described for *L. cuprina* resulted in a gene fragment of ~200 bp containing the site of the Gly137®Asp mutation. This region showed high similarity to the orthologous region in *L.*cuprina. The intron position is conserved, however its physical size and nucleotide sequence are not. The partial exon sequence differed from that in *L.cuprina* at 2 nucleotide sites, but no amino acid substitution was predicted. Further analysis will be conducted to determine the association of this point mutation OP resistance in *C.hominivorax* strains. This characterization should allow the identification of resistant individuals, enabling more effective control strategies.

Evaluation of prepared Thai plant-based repellents against mosquito bites under field condition

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The experimental repellent products in the gel dosage form contained 20% clove oil (*Syzygium aromaticum*) and 10% clove plus 10% makaen (*Zanthoxylum limonella*) oil mixture were evaluated theirs efficacy against daytime- and nighttime-biting mosquito comparing with 20% deet (di-methyl benzamide) under field condition using a human bait method. One gram of the product was applied evenly from knee to the ankle of each leg. Shorts and shoes were worn to standardize the exposure area. Other exposed untreated parts of the body were protected against mosquitoes attack by wearing a jacket with hood and gloves, covering the thighs with a plastic sheet. For daytime-biting, 20% clove showed complete protection for 4.0 h and gave 95.7% repellency after 5 h application, whereas 10% clove plus 10% makaen oil mixture and 20% deet provided only 86.8 and 82.7% repellency after treatment, respectively against *Aedes aegypti*. For nighttime-biting, the 3 developing repellents yielded equally excellent (average 97.1% repellency for 5 h against the predominant *Culex quinquefasciatus* and *Mansonia uniformis* but they gave 89.0% repellency against *Cx. tritaeniorhynclus* and *Cx. gelidus*. This finding demonstrates that the repellency of 20% clove oil and 10% clove plus 10% makaen oil mixture in the gel dosage form are possible used against various different mosquito species.

Host grooming efficiency and its influence on the distribution of cat fleas, *Ctenocephalides felis*

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Grooming is usually supposed to be the main factor to affect the final distribution of ectoparasites on hosts, that is, the distribution of ectoparasites can be changed when the grooming ability is refrained or can be different between those on the good and poor groomers. Therefore, in this study we compared the distribution of cat fleas, *Ctenocephalides felis* (Bouché)(Siphonaptera: Pulicidae), on the cat with good or poor ability of grooming and the cat wearing an Elizabethan collar or not. As expected, the results show that cat's grooming efficiency against fleas was significantly varied among individuals and greatly reduced by wearing an Elizabethan collar. However, the effect of grooming efficiency on the distribution of fleas on the host body is not significant. Regardless of cat's grooming efficiency, mean percentages of fleas detected at 24 or 96 hours after initial infestation were always highest on the neck (52 – 71.67%) among body regions (other regions ranging from zero to 30%). Fleas seem to prefer the anterior regions of cats including the head, neck, and front back. Moreover, male (60.17%) and female (61.18%) fleas have the same trend to move to the cat's neck.

Impact of anthelmintics on the growth of dung beetles and dung breeding flies in Japan.

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Most of anthelmintrics like an Ivermectin is excreted in the feces as a mixture of parent compound and metabolites. So, the use of anthelmintrics in livestock has potential impact on nontarget invertebrates, for example, dung beetles, flies, that depend on dung pats and contribute dung degradation in grassland. This study examined the impact of the anthelmintics residue on the dung degrading fauna in grassland. Dung samples were collected on 1, 3, 7, 14, 21, 28 and 35 day after pour on and bioassayed using the dung beetles and flies. Ivermectin residues attained maximal concentration from 1st to 3day after pour on followed by a linear decline to 35day. Dung pats collected 1 to 14days after treatment with ivermectin were lethal to larval development of native dung beetle, *Onthophagus lenzii* and native dung breeding flies, *Musca hervei*. Dung collected 1 to 3days after treatment was lethal to larval development and pupation of native dung beetle, *Aphodius elegance*. In field experiment using emergence trap, number of emerged flies were 3,655 (18families) in control dung and 2,234 (16 families) in treated dung.

Investigation of alternative insecticides for the treatment of bed nets and for indoor residual spraying to control *Anopheles* mosquitoes.

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There is an urgent need to investigate alternative insecticides for the control of pyrethroid resistant *Anopheles* mosquitoes, vectors of malaria. Resistance to DDT and pyrethroids among *Anopheles* (especially *gambiae* and *funestus*) species has been reported in western, southern, and eastern Africa. Alternative insecticides with novel modes of action remain may have potential for use on nets or indoor residual sprays. The Gates Malaria Partnership at LSHTM is working with WHO and industry to identify new actives. Bioassay tests were carried out using new insecticides including a phenylpyrazole (fipronil), an organophosphate (chlorpyrifos methyl, "Reldan"), an oxadiazine (indoxacarb) and a pyrrole (chlorfenapyr). Some of these compounds were tested in mixtures to investigate their possible synergistic interaction with pyrethroid insecticides. Laboratory studies with fipronil on netting have shown some irritant effect against *An. stephensi* and some synergisms between fipronil and deltamethrin. Different formulations of Reldan sprayed on various substrates (mud, plaster and wood) to investigate the efficacy of the formulations for indoor residual spraying (IRS) showed good residual performance up to 6 months especially on plaster. On the treated nets, Reldan performed well against wild populations of pyrethroid resistant mosquitoes. Indoxacarb and chlorfenapyr showed a slow acting effect in terms of mortality and KD but indoxacarb was better on netting when mixed with deltamethrin.

Evidence supports two cytologically different races of *Anopheles pullus* in Korea

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Despite the medically-importance of anopheline mosquitoes as vectors of Korean vivax malaria, differentiation among Korean anopheline mosquitoes are obscure and difficult by traditional morphological taxonomic criteria. To verify the species status of two ambiguous species of *An. pullus* and *An. yatsushiroensis* (*An. pullus form yatsushiroensis*) mosquitoes from Korean, we established isofemale lines and investigated their genetic relationship by metaphase karyotypes, hybridization experiments, DNA sequence comparisons of rDNA ITS2 and mitochondrial COI and COII. Two isofemale lines showed different chromosomal shapes in sex chromosomes of X and Y, the reciprocal crosses between the two isofemale lines, however, provided viable progeny with completely synaptic salivary gland polytene chromosomes. DNA analyses also strongly supported the conspecificity of these isofemale lines. Based on these evidences, we conclude that two *An. pullus* forms which differ distinctively in morphological, cytological and ecological traits remain in interfertile status.

Incompatible hybridization between two sympatric species of *Anopheles sinensis* and *Anopheles pullus* (Diptera: Culicidae) in Korea.

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Hybridization tests of laboratory-raised, iso-female lines of *Anopheles sinensis* Form B and *An. pullus* Form A and B were conducted by induced copulation. The three iso-females line were established based on morphological characteristics of wild-caught, fully engorged females and three distinct types of metaphase karyotypes. They were *An. sinensis* form B (X, Y_2), *An. pullus* form A (X_1,Y_1) and form B (X_2,Y_2). The results of reciprocal and back crosses among *An. sinensis* and *An. pullus* indicated that *An. sinensis* was genetically incompatible with *An. pullus*, providing low viability of F_1-hybrid larvae with completely asynaptic of salivary gland polytene chromosomes, sex distortion of adults, abnormal development of ovarian follicles of F_1-hybrid females, and atrophy of accessory glands and testes of F1-hybrid males. The results strongly suggested that these two morphological species exhibited entirely reproductive isolation.

Tick & tick borne diseases: antigen selection in a genomic era.

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Paper not available at time of production.

Reproduction & sex ratios in the lice of humans & animals

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Paper not available at time of production.

Attractiveness and food value of plants for aphids

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Plants of chrysanthemum, pink and cineraria were grown in greenhouse under either normal composition of elements (nitrogen, phosphorus, potassium), excess (+N,+P,+K) or shortage (-N,-P,-K) of one of these elements. Attractiveness of plants for aphids *Coloradoa rufomaculata* Wils. and *Myzodes persicae* Sulz. was determined as a number of adults, which occupied the plants. All the plants had maximal attractiveness at +N treatment, and the minimal at -N. The excess of the other elements resulted in increase in attractiveness compared to normal composition, the shortage led to decrease of attractiveness. Food value of plants was determined by fecundity, periods of development and reproduction, and longevity of the aphids. Fecundity was maximal under +K and especially +P, it was minimal under shortage of these elements. Treatments +N and -N did not differ from control and from each other. Plants under +P contained maximum of free amino acids. Longevity and reproduction period were increased under -N, however no difference between other treatments was observed. Development time did not depend on the treatments. Therefore the plants +N most attractive for aphids were less favourable for their population.

Ludwigia **flowers (Onagraceae): a rich food source for solitary bees in South Brazil**

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Species in the genus *Ludwigia* (Onagraceae) probably originated in wet habitats of the tropics of South America, being this area considered the centre of diversity for the genus, and containing the most primitive species. There are approximately 80 species of *Ludwigia* that occurs predominantly in the Neotropics. The habitat of *Ludwigia* includes swamps, marshes and wet places, being therefore common found in the lowlands of the Brazilian territory. The flower visitors of *Ludwigia* species were sampled periodically in southern Brazil (Santa Catarina and Rio Grande do Sul - 28° to 30°'S), where the coast plain is broad and with big freshwater lagoons from the Quaternary. The insects were sampled with entomological net and observed during the visits. More then 30 bee species were identified as frequent visitors of the *Ludwigia* flowers. Furthermore, at least 16 species were considered oligolectic to these plants, which means that they are specialized to collect pollen from one or more species of *Luwigia*. In other words the female and the larvae of the bee depend on the floral resources of the *Ludwigia* species. These plants can be considered important components of the melittophilous vegetation since they support around 10% of the local bee fauna.

Applying GIS for finding distribution patterns of arboreal Coleoptera: cultivating a seed program for inventories and conservation planning in western Amazonian forests.

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Over the last decades, descriptions of tropical forests beetles have made a large taxonomic contribution in Coleoptera, but the mechanisms and rules explaining the species distribution and abundance in the tropics are still poorly understood. Data collected from a canopy arthropod biodiversity study conducted in Ecuadorian Amazonian forests between January 1994 and July 2002 is here used for exploring spatial and temporal distribution patterns of beetles responding to tree species occurrence and microhabitat characteristics. Point maps of beetle distributions were produced at a scale of meters using ArcViewGIS and the emergent distribution pattern was tested against canopy variables, such as: tree species composition; strata structure; occurrence of dry leaves, lianas and epiphytes, and light measures. Using GIS techniques was justified by the high diversity and complexity of Amazonian forests. Distribution patterns of beetles at a fine scale responding to forest composition and structure should reflect animal–plant ecological relations, so our results set a starting point in developing further studies which will seek to understand functioning of the richest biotic communities on Earth. Abundance distribution information for beetles at a very fine scale can also be directly applied in planning inventories studies and taking local conservation decisions.

Correlation of stylet penetration behaviours and an EPG waveform with inoculation of *Xylella fastidiosa* **by the glassy-winged sharpshooter,** *Homalodisca coagulata*

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Stylet penetration behaviours of the glassy-winged sharpshooter, *Homalodisca coagulata* Say, are being studied to determine the behavioural mechanism of inoculation of *Xylella fastidiosa* (Xf) in plants. To do this, we are using AC and DC electrical penetration graph (EPG) monitoring of feeding, videomicrography of stylet movements, and histology of both salivary sheaths in plants and Xf aggregations in vector precibaria. We report herein correlations of GWSS AC waveforms A1 and A2 with salivary sheath formation, B1 and B2 with other pathway activities, and C with excretory oplets (presumed ingestion). Results presented also implicate the B1 waveform in inoculation of Xf near identified probes, and discuss whether bacterial populations in the vector precibarium can be correlated with bacterial expulsion and/or any waveforms. The ultimate goal of our research is to develop a Stylet Penetration Index using EPG monitoring of feeding, which could provide a rapid means of screening grape varieties for host plant resistance to the vector.

Correlations of probing behaviour by glassy-winged sharpshooter, *Homalodisca coagulate* **(Say), with AC electrical penetration graph (EPG) waveforms using video and artificial diets**

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Glassy-winged sharpshooter (GWSS), *Homalodisca coagulate* (Say), is the main vector of *Xylella fastidiosa,* the casual bacterium of Pierce's Disease in grapevine. A detailed understanding of GWSS probing behaviour is important because *X. fastidiosa* is acquired and inoculated when GWSS feeds. To further elucidate GWSS probing behaviour we used the AC electrical penetration graph technique (AC EPG) and video to correlate GWSS probing behaviours with EPG waveforms in artificial diets. Three previously described waveforms, A1, B1 and B2, were correlated with GWSS stylet activity. Fascicle protraction, alternating mandible retraction and maxillary tip fluttering were correlated with subdivisions of the A1 waveform. Salivary sheath formation was also observed in A1. Waveform B1 was correlated to maxillary fluttering within the salivary sheath. Termination of the B1 waveform and maxillary fluttering would sometimes occur when the maxillae were inside the saliva sheath and always when the maxillae exited the salivary sheath. Waveform B2 was associated with stylet path branching within the salivary sheath. During B2, GWSS would "saw" a new stylet branch in the salivary sheath by rapidly retracting and protracting its stylet fascicle in an eschewed direction from the original branch. Termination of B2 occurred when the stylets "sawed" through the salivary sheath.

Investigations on possible plant factors which may contribute to the spatial distribution pattern of aphid species living on tansy (*Tanacetum vulgare* L.)

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Tanacetum vulgare L. (Asteraceae) is the host plant for a guild of aphid species, all feeding on the phloem, but utilizing different feeding niches, for example fresh shoots, senescing leaves or the root collar. The aim of this study was to investigate if plant morphological and physiological factors, i.e. food accessibility and its nutritional quality in different plant parts, contribute to the aphid species' feeding site preferences. Light microscopical measurements of the distance between epidermis and phloem tissue revealed that food accessibility in most plant parts was not restricted by the stylet length of the examined aphids. Phloem nitrogen content in form of free amino acids is hypothesised to affect aphid performance and acceptance of host plants significantly. The amino acid composition of plant tissue extracts, aphid honeydew, as a waste product of the ingested phloem sap, and exudates from excised leaves in EDTA solution were determined. Samples derived from young, mature and old leaves were analysed. Preliminary results suggest similar within-plant amino acid profiles while inter-plant patterns may vary considerably.

Evaluation of R genes for durability of gall midge resistance in rice

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The Asian rice gall midge (*Orseolia oryzae)* is a serious pest of rice in several Asian countries. Development and deployment of resistant varieties has been the key approach for tackling the pest menace. However, rapid development of new virulent biotypes of the pest breaking down the resistance has been the main limitation. One of the breeding strategies now being adopted is to pyramid two or more genes in the same cultivar through marker assisted selection. The critical question now is to know which gene combinations are likely to provide desired durability. We have been addressing this question through several different approaches. Through a simple method to test the virulence spectrum of individual mated female of the rice gall midge *Orseolia oryzae,* we have noted changing virulence through time and space across several pest endemic locations. Through studies on inheritance of virulence it is observed that virulence acquired through the sex linked recessive gene (*vGm2*) tends to be less durable compared the somatic recessive gene (*vGm1*). Fitness cost associated with virulence against *Gm1* gene may be responsible for the observed unstable virulence. Some of the gene combinations when tested under multi-location testing have displayed potentiating effects, while other combinations are just additive.

Symbiotic control of Pierce's disease: biology of mutualistic bacteria in grapevines and bioassay of reagents to disrupt pathogen transmission

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The grape strain of *Xylella fastidiosa* (*Xf*) causes Pierce's disease (PD) in grapevines and is transmitted by sharpshooter insects. The most efficient vector for transmission is the glassy-winged sharpshooter (GWSS), *Homalodisca coagulata*. Symbiotic control employs mutualistic bacteria to deliver anti-Xf compounds to disrupt transmission of the pathogen to new host plants. *Alcaligenes xylosoxidans* var. *denitrificans* (*Axd*) is a prime candidate for control of PD because it inhabits the foregut of GWSS and the xylem of plants and has access to the pathogen, *Xf*. Registration of Axd as a microbial pesticide requires the ecology of *Axd* to be determined along with any side-effects. Candidate toxins for use in *Axd* have disrupted disease transmission. *Axd* colonized and travelled within 6 host plants, reaching the highest titers in lemon. *Axd* colonized the foregut region of two alternate *Xf*-vectors, the blue green and smoke tree sharpshooters, and was not identified in predatory arthropods that fed on *Axd*-positive GWSS. *Axd* genetically modified with a fluorescent marker gene did not contaminate the berries of grapevines in commercial vineyards.

An ecological-based approach to drug discovery using plant-insect interactions

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Ecological-based approaches to drug discovery use plant-herbivore theories to make predictions about secondary metabolite distributions within the plant kingdom. The ecological-based approach used here models an aspect of the plant apparency/plant predictability theories, and predicts that herbivores select for a diverse array of plant chemicals. Based on the feeding patterns of 32 specialized and generalized herbivores, we predicted that deciduous woody perennial shrubs and trees would be the most likely plant growth form to produce a diverse group of chemicals effective against human infectious diseases. Test results from 63 Sonoran Desert plant species indicated that woody perennials and several long-lived herbaceous perennials were the most inhibitory to *Staphylococcus aureus* and *Candida albicans* when compared to the annual and evergreen life forms. Of these 63 plant species, 31 were used by indigenous peoples to treat infectious diseases, and 32 were randomly selected. In both cases about 44% of the species were active against the above microbes. Overall, the results indicate that plant-herbivore interactions can be used to enhance the opportunity to find extracts effective against human diseases.

Is bluegreen aphid resistance in *Medicago truncatula* induced?

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Bluegreen aphid (*Acyrthosiphon kondo*) survival and growth is reduced on the aphid-resistant (R) *Medicago truncatula* cultivar Jester compared to the near-isogenic susceptible (S) cultivar Jemalong. Electrical penetration graph (EPG) assays have indicated reduced phloem ingestion on Jester, suggesting phloem-based resistance. This study further characterise the resistance by assessment at the whole plant or tissue level, and examining the effect of prior aphid exposure on feeding behaviour. Performance of nymphs was assessed on shoot tips from both Jester (R) and Jemalong (S), either on the plant or excised from the plant and maintained on supplemented agar. Fecundity, survivorship and colony growth was reduced ($P<0.05$) on the Jester (R) compared to Jemalong (S), however no differences were observed between the excised tips from these same cultivars. This suggests at least one factor is absent from the excised tissue that is necessary for resistance to be manifested. Feeding behaviour of individual aphids was assessed using EPG on previously aphid-infested vs. uninfested plants of Jester (R) and Jemalong (S). Phloem ingestion was significantly reduced on the resistant Jester plants, confirming previous studies. Phloem ingestion on previously infested plants of both Jester and Jemalong was also reduced, indicating systemic activation of a phloem specific resistance.

Integrating optimal foraging and optimal oviposition theory in plant–insect research

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Most research on host selection by arthropods is based on optimal oviposition theory. This theory, also called the oviposition preference–offspring performance hypothesis, predicts that a female will choose those hosts for oviposition on which larvae perform best. Yet, many phytophagous insects do not only use plants for larval feeding but also as adult resources. As a consequence, optimal foraging by adults may also shape host preference. This hypothesis predicts that adults will prefer the host(s) optimal for their own performance (= realised fecundity). We show that at least in three phytophagous insects, there exists strong evidence that adult females feed and oviposit on the hosts that are optimal for adult nutrition rather than on those optimal for offspring development. Therefore and because optimal foraging and optimal oviposition may oppose conflicting needs to phytophagous insects, we plea for an integration of optimal foraging and optimal oviposition in plant–insect research.

Temporal variations in the community of galling insects associated to *Baccharis acunculifolia* (Asteraceae): effects of weather, resource abundance, plant sex and gall competition.

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This study aimed to verify the effects multiple factors on the abundance of three galling insect guilds (stem, leaf and bud galls) supported by the dioecious *Baccharis acunculifolia* throughout one year. Five plant traits were sampled: number of fourth-level shoots, shoot growth rate and number of ramifications, leaves, and inflorescences. The effects of weather and plant traits on temporal variations in the abundance of each guild were analysed, as well as the interactions between gall species and differences in plant traits and gall abundance between male and female plants. *Baccharis acunculifolia* supported 14 insect gall species. This gall community was characterized by a strong dominance of the leaf galling psyllid *Baccharopelma acunculifoliae*, which accounted for 80% of gall occurrences. The weather did not affect gall abundance directly, but influenced the number of fourth-level shoots. This variable indicates the amount of plant meristems, a fundamental tissue to gall induction. The abundance of *B. acunculifoliae* negatively affected the abundance of other galling species, possibly indicating interspecific competition. There were no intersexual differences in plant traits and gall abundance. As a whole, these results corroborated the "resource abundance hypothesis", since gall abundance was affected by the amount of resources offered by *B. acunculifolia*.

The sensory receptors of the labio-maxillar complex of larvae in Colorado potato beetle, *Leptinotarsa decemlineata* Say (Coleoptera: Chrysomelidae)

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The Colorado potato beetle, *Leptinotarsa decemlineata* Say, is a devastating pest of potatoes and other solanaceous crops. The types, numbers and distribution of sensory receptors on the labio-maxillar complex of larvae were studied with the scanning electron microscopy. A total of 32 sensilla 3 basic morphological types - basiconic, trichoid, and digitiform are present on the maxillary and labial palps. The labial palp has 11basiconic sensillae. All this sensilla are placed on the distal apex of palp. The length mean of basiconic sensillae were 29 micron. The maxillary palp has 16 basiconic, 4 trichoid and 1 digitiform sensilla. All basiconic sensilla are placed on the distal apex of the third segment. The length mean of basiconica sensilla were 31.2 micron. Also were observed 2 morphological types of basiconic sensilla. In other word, placed 3 trichoid sensilla on the distal of the second segment and 1 trichoid sensillae on the lateral side of third segment. Also 1 digitiform sensillae were observed on the lateral side of third segment of maxillary palp. The length and wide of these sensilla were recorded 16.5 and 3 micron, respectively. The percentage composition of the types of receptors present: basiconica 84.4%, trichoid 12.5% and digitiform 3.1%. The work was supported by RFBR (grant # 04-04-48779).

Life history of ***Macrosiphum euphorbiae*** **on tomato plants previously infested with** ***Bemisia tabaci***

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Macrosiphum euphorbiae and *Bemisia tabaci* are two important pests of tomato crops and efficient vectors of plant viruses. Induced resistance (IR) is a promising mechanism for crop protection that offers a long lasting defence against a broad spectrum of diseases in various plant species. IR may be elicited by previous infestation by other herbivores. However, host plant induced responses are not necessarily negative and could produce beneficial effects on other insect pests. We evaluated the effect of a previous infestation by *B. tabaci* on the life history of *M. euphorbiae* on tomato plants in laboratory conditions for 48h. Two alate aphids were put on these tomato plants during 24h and removed, leaving two nymphs per plant, born during this period. We evaluated the offspring after 10 and 20 days, i.e. the total number of nymphs and adult aphids per plant. Our results indicate that the population of *M. euphorbiae* increased to a greater extend when tomato plants had been infested previously with *B. tabaci* than if not previously infested.

Genetic and biochemical differences in host-plant use by the aphid *Myzus persicae* (Sulzer)

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Specialization in phytophagous insects is mainly mediated by a differential adaptation to the host's allelochemicals. Aphids are ubiquitous sap-sucking insects that frequently constitute agronomical pests. One of the most generalist aphid species is *Myzus persicae*. However, *M. persicae* has developed a particularly well adapted subspecies (*M. persicae nicotianae*) on tobacco (*Nicotiana tabacum* L.), that exceptionally has been detected on other hosts. This high level of specialization must comprise a genetic/biochemical base. Seventy-six aphids were sampled from different host-plants in Central Chile. We characterized 14 generalist genotypes and one single genotype on tobacco using microsatellite markers. Biochemical and genetic differences were compared between a generalist genotype reared on pepper (*Capsicum annuun* L.) and the single genotype specialized on tobacco. Significant differences in activities of the detoxification enzymes cytochrome *P*-450, general esterases, and glutathione *S*-transferase between genotypes were found. The tobacco-specialized genotype on tobacco showed the highest activity for cyt *P*-450, but the lowest for the other enzymes in comparison to the generalist genotype. We are investigating if those results are due to differences in genotypes or in gene expression.

Ultrastructural contributions to the study of the glassy-winged sharpshooter feeding and Pierce's disease.

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The glassy-winged sharpshooter (GWSS) is a vector for the transmission of the bacteria *Xylella* fastidiosa, which causes Pierce's Disease in grapes, leaf scorch in oleander and almonds, variegated chlorosis in citrus as well as causing diseases in avocados, peaches, plums, apricot, cherries, alfalfa, and many trees and ornamentals. This xylophagous leafhopper feeds on more than 100 different host plants from more than 35 plant families. Recent ultra structural studies have contributed significantly to our understanding of sharpshooter feeding. Our continuing studies on sharpshooter feeding have elucidated the damage that the stylets can cause to host plant tissues during the penetration process. The penetrating stylets damage the cells of the plant host on the route from the epidermis to the xylem tissue. Examination of the walls of vessel elements show that they are often shattered during GWSS stylet penetration. The salivary sheaths formed by the sharpshooter during penetration and feeding may be simple or highly branched and terminate in parenchymatous or vascular tissues of the host plant. Salivary sheath material was also observed to occlude the lumina of the vessel elements, presumably preventing the normal translocation of xylem fluid.

Does voltinism in temperate insect herbivores depend on defences of their host plant? Comparative analysis using European butterflies

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According to theories of plant antiherbivore defences, leaf-chewing insects feeding on apparent plants should synchronise hatching with appearance of fresh leaves. In contrast, such a synchronisation should be less pronounced in insects feeding on unapparent, qualitatively protected plants. We hypothesised that in seasonal temperate environments, quantitative plant defences should constrain the annual number of insect generations. Testing the hypothesis using European butterflies corroborated the prediction. Apparency in a broader sense (woody plus gramineous plants) performed as a strong predictor of low generation numbers; when controlled for phylogeny, even a narrower definition of apparency (woody plants) predicted low number of generations. Other significant predictors of monovoltinism were living in arctic/alpine environments and having large bodies; large body size was a weaker predictor than host apparency. The nature of host plant defences may explain several little understood patterns in phenology of temperate insects, including scarcity of late-season monovoltines, and a high numbers of generations in insects associated with ruderal plants.

Transmission of citrus endophytic bacterium *Methylobacterium* by sharpshooters (Insecta: Cicadellinae): vectors of *Xylella fastidiosa*, the causal agent of variegated chlorosis of citrus.

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Endophytic bacteria are those that can be isolated from surface disinfected plant tissues and have no deleterious effects on the host plant. These microorganisms have shown potential benefits as biocontrol agents of pests and diseases. The Citrus Variegated Chlorosis is caused by the bacterium *Xylella fastidiosa,* which is transmitted from infected to healthy plants by sharpshooters: *Dilobopterus costalimai, Oncometopia facialis* and *Acrogonia* sp. In citrus plants, *X. fastidiosa* and endophytic *Methylobacterium* may colonize xylem vessels, allowing endophyte transmission by the same way of pathogenic bacteria. Trying to elucidate this point, bacteria were isolated from heads of insect vectors and citrus plants. *Methylobacterium* was the most frequent bacterial specie in both situations and shown *in vitro* interaction with *X. fastidiosa*. *Methylobacterium* expressing GFP gene was obtained, and further used to study the transmission of this bacterium by sharpshooters. The insects were fed in membrane with the GFP bacteria and than trapped in *Catharanthus*, a model plant. After 40 days, plants were surface disinfected and macerated to isolate bacteria. It was observed that transmission of the GFP bacteria occurred in 13.3% of the plants, showing that the endophyte is being transmitted to plants by the same vector of *X. fastidiosa*.

Discrimination of host plants by *Rhopalomyia* species at various levels of botanical variation

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Most gall midges (Diptera: Cecidomyiidae) are monophagous or oligophagous, although a few are polyphagous and three species exhibit host alternation. This suggests that speciation of gall midges may have occurred in association with evolutionary processes of plant species. It is an interesting study subject to investigate whether or not gall midges can discriminate, in the course of host range expansion, differences among their host plant individuals or populations. In this study, we examined morphological and molecular information on all 11 nominal Japanese *Rhopalomyia* species that induce galls on several species of the genus *Artemisia* (Asteraceae). We found that some of the Japanese *Rhopalomyia* species could not discriminate differences in the polyploidy among their host plant individuals. In six species out of the 11, the host range did not expand beyond the botanical section and no gall midge had a host range beyond different botanical genera. This may indicate that the gall midges cannot recognize the early stage of plant speciation and they expand their host range without knowing the genetic changes in plant species. Plant speciation that is initiated by chromosomal changes may provide associated gall midges with a chance to be isolated geographically or chronologically in the future.

Dirty cropping and insect community structure: can relaxed weed control benefit pest control in Brassicaceae crops?

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To assess the impact of increased vegetational diversity and complexity on insect abundance, mechanical weed control was relaxed on four organic farms in western Washington. By using commercially producing farms and production level plot sizes for Brassicaceae crops, I tested the feasibility of using relaxed weed control (dirty cropping) as a cultural pest control strategy. The four farms varied in the degree of implementing relaxed weed control, resulting in different densities of weeds in the dirty plots. The importance of how much weed control was practiced depended on which pest species was examined. Adult flea beetles (*Phyllotreta cruciferae*, and *Phyllotreta striolata* Chrysomelidae: Alticidae) were influenced at all levels of relaxed weed control. A suite of lepidopteran pests (*Pieris rapae*: Pieridae, *Plutella xylostella*: Plutellidae and *Trichoplusia ni*: Noctuidae) were not affected by any commercially viable level of relaxed weed control. Aphids (*Brevicoryne brassicae, Myzus persicae* and *Lipaphis erysimi* (Kaltenbach): Homoptera: Aphididae) had a slight dependence on the degree of relaxed weed control. Possible behavioural mechanisms and impacts of natural enemies are discussed to explain the observed results. The impacts on yield weight and quality for broccoli are presented for two of the farms.

Association between the deodar weevil *Pissodes nemorensis* (Coleoptera: Curculionidae) and the pitch canker fungus *Fusarium circinatum* in South Africa

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The deodar weevil *Pissodes nemorensis* is an important pest of pines, widely distributed in South Africa. The pitch canker fungus *Fusarium circinatum* is a serious pathogen in South African forestry nurseries, but has not been found on mature trees, possibly due to the lack of an effective insect associate. A Potential association between *P. nemorensis* and this pathogen is considered a serious threat. Greenhouse experiments were conducted where Fusarium-free weevils, weevils contaminated with *F. circinatum*, direct inoculation of *F. circinatum* into wounds and untreated controls were included. *Pinus patula* trees exposed to Fusarium-treated weevils showed dieback after six weeks while symptoms on *P. elliottii* were observed after ten weeks. All *P. patula* and *P. elliottii* trees had died after ten and twelve weeks respectively. Trees inoculated directly died more slowly than those exposed to Fusarium-infected weevils. No symptoms were seen on control trees. These results elevate concerns regarding a future association between *P. nemorensis* and *F. circinatum* in South Africa.

Globular structure and its morphological changes in mycangia of *Platypus quericivorus* (Coleoptera: Platypodidae).

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Platypus quercivorus is a vector of a pathogenic fungi *Raffaelea quercivora*, causal agent of Japanese oak disease. Females of *P. quercivorus* have about ten mycangial pit on their pronotum that is the storage organ for ambrosia fungi. In this study, globular structure was observed in each mycangial pit. This structure is consisted of several cells with thick cell wall covered by thick outer wall, and is almost large enough to occupy the space in its pit. We observed the temporal changes of the structure in mycangia from female enter the new gallery. We inoculated female beetle into logs that have been killed and inoculated male in advance. Twelve hours after the female inoculation, the outer wall of the globular structure partly broke, and after twenty fore hours, each cell inside became into a lump of yeast-like smaller cells in several pits of each individual. No globular structure was observed any more in the pits with yeast-like cells. Secretory cells around the mycangia were thick for these periods. Then the contents in all mycangial pits were a lump of yeast-like cells two days after inoculation, and secretory cells have been thin.

The origins of olfactory preference in an aphid parasitoid

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Aphidius colemani (Viereck) parasitoids emerging from peach-potato aphids, [*Myzus persicae* (Sulzer)] reared entirely on plants of the Brussels sprout variety Winter Harvest (*Brassica oleracea* L. var. *gemmifera* Zenker) were tested for odour preference in an olfactometer. When parasitoids were allowed to choose between a Brussels sprout plant odour and clean air, they preferred the plant odour irrespective of whether that variety was the host aphid's food plant. However, when the parasitoids were tested against two different varieties of Brussels sprout they consistently preferred the host's food plant (Winter Harvest). This preference was retained even when they eclosed from mummies isolated from the plants or when the parasitoid pupa was surgically removed from the mummy case, prior to emergence. We are presently examining the origin of preference using aphid hosts reared on an artificial, plant-free, diet.

Evolution of gall induction in *Lachnodius* Maskell (Hemiptera: Coccoidea: Eriococcidae)

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Lachnodius, interesting in terms of its aberrant cytogenetics, is one of a handful of eriococcid genera inducing galls on eucalypts in Australia. In comparison with these other genera, *Lachnodius* is unusual in containing species that are both free-living and that induce a variety of galls ranging from shallow pits to more elaborate, enclosing structures. We estimate relationships among *Lachnodius* species and some of their relatives from a phylogenetic analysis of the morphology of first-instar, second-instar, and adult females. The evolution of gall-induction is explored by mapping gall traits onto the consensus tree. The free-living species of *Lachnodius* are sister to the gall-inducing species. Multiple transitions from the simple pit-gall form to the superficially more complicated enclosing galls have occurred. *Lachnodius* is paraphyletic with respect to both *Opisthoscelis* Schrader and *Sphaerococcopsis* Cockerell, in which adult females exhibit an extreme bias towards the development of the hind legs.

Root application of b-aminobutyric acid (BABA) inhibits growth and reproduction of the pea aphid (*Acyrthosiphon pisum*) on legumes.

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b-aminobutyric acid (BABA) is a non-protein amino acid that is an effective inducer of resistance against a variety of plant pathogens. However, examples of BABA-induced resistance against insect herbivores are scarce. We applied BABA as a soil drench to various legumes and monitored its effect on the pea aphid (*Acyrthosiphon pisum* (Harris)). On tic beans (*Vicia faba* v. *minor*) BABA caused a dose-related reduction in the mean relative growth rate (MRGR) of individual aphids and also in the intrinsic rate of population increase (r_m). BABA (25ml of 50mM solution per plant) also caused significant reductions in MRGR of *A. pisum* on peas (*Pisum sativa*), broad beans (*Vicia faba* v. *major*), runner beans (*Phaseolus coccineus*) and red clover (*Trifolium pratense*). The concentrations of BABA used in our assays were high compared to those used to induce resistance against phytopathogens such as viruses, bacteria and fungi. It is possible that the reduction in aphid performance was due to a BABA-induced phytotoxic stress-response in the plant, although no obvious reductions in plant health were observed. Alternatively, BABA may exhibit direct toxicity against aphids. Current experiments are investigating the mechanisms by which BABA reduces aphid performance.

The role of indigenous pollinators in New Zealand onion crops.

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Arthropods are important vectors for pollen transfer in many cropping systems. Their movement and effectiveness as pollinators have major implications for pollination services, contamination of certified seed crops and for the coexistence of GE and non-GE crops. To assess the potential risk of gene escape from transgenic crop plants, pollen mediated gene flow needs to be estimated. This requires an evaluation of pollinator assemblage diversity and the ability of vectors to move viable pollen over distance. *Allium* crops across New Zealand have been chosen as one model system for a multi-site, multi-year assessment of variation in pollinator assemblages and abundance. Assemblage differences due to seasonal influences and environmental factors are being examined as they can can produce uncertainty in risk assessments. Data from the first year of results will be presented to illustrate the diversity of Arthropods carrying conspecific pollen within onion fields.

Fungus gnats and their possible association with the pitch canker fungus in South African pine nurseries

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Fusarium circinatum is the causal agent of a serious disease of seedlings in South African pine nurseries. Insects and especially fungus gnats (Diptera: Sciaridae, Mycetophlidae) were suspected of transmitting this fungus in nurseries. The aim of this study was to ascertain which species of gnats are present in South African pine nurseries, and to consider whether these and other Diptera carry *F. circinatum*. Adult Diptera were surveyed in four major forestry nurseries between 2000 and 2001. Fungi were isolated from these insects and resulting *Fusarium* spp. were identified. *Bradysia difformis* was the only species of fungus gnat found and it occurred in all nurseries that were surveyed. This is the first report of *B. difformis* in South Africa. Other dipteran families collected included Agromyzidae, Cecidomyiidae, Chironomidae, Ephyidae, Muscidae, Simulidae and Tachinidae. Of these, only Ephyidae has a history of interaction with plant pathogens. *Fusarium circinatum* was not isolated from any of the Diptera. *Fusarium oxysporum* and *F. stilboides* were isolated from Chironomidae, but these fungi are not considered important pathogens of pines in the nurseries surveyed.

Effect of cultivars of cabbage *Brassica oleracea* var *capitata* in the biology of *Plutella xylostella* (L. 1758) (Lepidoptera: Plutellidae)

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This research proposed to study the effect of eight cultivars of cabbage subordinate to *Plutella xylostella* attack, in two successive generations. The duration and viability of the larval phasis, the duration and viability of the pupa phasis, pupa's weight in 24h of life, period of pre-emplacement of the eggs, sexual reason, number of eggs per female, period of hatching and viability of eggs were evaluated in the two generations. The corrected reproductive potential was calculated. The experimental delineation was completely casualized, in factorial scheme of eight cultivars and two generations, with five repetitions. The results were submitted to an analysis of variance (Schefer Test) and the averages were compared by test of Tukey (a =0,05). The cultivars of cabbages that were evaluated, affected the biology in the *P. xylostella*, being Ruby Ball, Bull's Heart and Matsukase Sakata moderately resistant, while, Mad of Summer, Flat of Yard and Red Giant Mamuth and Toshin Takii were susceptible, and the Sixty Days was highly susceptible to *P. xylostella*. Matsukase Sakata and Ruby Ball were those ones that most affected the corrected reproductive potential of the plague.

Insecticidal aqueous vegetable extracts effects against *Ascia monuste orseis* **at kale**

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The aim of this work was to study the aqueous extracts effects of *Azadirachta indica* almonds and *Sapindus saponaria* fruits against *Ascia monuste orseis.* The CL_{50} estimated for larvae of first instar were 0.0117% (w/v) and 1.0342% (w/v) for *A. indica* and *S. saponaria* extracts, respectively. The lethal concentration for *A. indica* and *S. saponaria* were respectively 0.0858% and 6.8826%. The CL_{50} for *A. indica* and *S. saponaria* extracts with the addition of adhesive spreading were 0.0133% and 1.0461%, respectively. Mortalities of the insects were similar with or without the use of adhesive spreading. The concentrations of aqueous vegetable extracts affected the insect biology. The extracts affected the eggs number oviposited and feeding of the *A. monuste orseis*, due to theirs repellents and/or deterrents effects and/or feeding suppressor presence. The extracts presented insecticidal action at the embryonic development and at recently hatched larvae with respective concentrations of 0.5% and 0.1% for *A. indica* and 3.0% and 6.0% for *S. saponaria.* The insecticidal effects were bigger at eggs treated with 48 hours of life than those eggs that had just oviposited. The results demonstrated that the aqueous extracts of *A. indica* almond and of *S. saponaria* fruits are promising for *A. monuste orseis* control.

Feeding behavior of the bean bug, *Riptortus clavatus* (Thunberg), on seeds of a resistant mungbean variety

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The bean bug, *Riptortus clavatus*, couldn't survive when supplied with seeds of a mungbean (*Vigna radiata* (L.) Wilczek) variety, Jangannokdu, though it normally developed on seeds of most other mungbean varieties. The feeding time of the fifth instar nymph per day on a resistant variety was not significantly different from that on a susceptible variety. In a preference test in a petri-dish, the number of puncture marks, stylet sheaths, formed on seeds was not significantly different between the two kinds of varieties. However, the feeding amount (ca. 1.9 mg) per day of the fifth nymph on a resistant variety was significantly less than that (ca. 4.5 mg) on a susceptible variety. The results, therefore, indicated that the bean bug couldn't feed on seeds of the resistant variety although it inserted its stylet into the seeds. When the coat-removed seeds were provided for the bean bug, the survival rate was very low to show that the seed-coat didn't have a resistant factor. When the bean bug was reared using artificial diets mixed with the seed powder of both varieties, the survivorship of the insect decreased, depending on the dose of the powder of a resistant variety.

Biochemical analysis on the induced resistance to pathogens in rice plants infested by *Sogatella furcifera*: accumulation of defense-related chemicals in rice plants, such as jasmonic acid, salicylic acid and phytoalexins.

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We reported that the rice plants previously been infested by the white-backed planthopper, *Sogatella furcifera*, strongly induce the systemic resistance to the rice blast fungus caused by *Magnaporthe grisea* (Kanno and Fujita, 2003). Moreover, we recently found that the rice plants infested by *S. furcifera* also acquired resistance to the bacterial blight caused by *Xanthomonas oryzae*.

To clarify the mechanism of such induced resistance, we analysed some defense-related chemicals, such as jasmonic acid, salicylic acid and phytoalexins (Momirakton-A and Sakuranetin) in rice plants using LC-MS. Accumulation of these chemicals in *S. furcifera* infested plants was higher than those in uninfested control plants. Especially, in *S. furcifera* infested plants, both phytoalexins, Momirakton-A and Sakuranetin, that indicate strong antimicrobial activity to the fungus were much more accumulated than that in uninfested control plants. These results suggest that the defence systems, such as jasmonic acid pathway and salicylic acid pathway, must be activated so that phytoalexins are biosynthesized in rice plants.

Inoculation of the adult beetles of *Platypus quercivorus* to Japanese oak trees

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The adult beetles of *Platypus quercivorus* were artificially inoculated to Japanese oak trees (*Quercus crispula*). Two inoculation methods were set, A) a high density inoculation with the pipette chips and B) an inoculation by release into the netting. Four of five trees which were inoculated by the pipette chips were dead, and five trees which were inoculated by the netting were all dead. Seven of nine dead trees were presented the same wilting symptom as the current mass mortality of oak, and withered completely within 1-2 weeks. *Raffaelea quercivora*, which was confirmed as the pathogenic fungus of the wilt disease and was usually isolated from mycangia of *P. quercivorus*, was isolated from all the inoculated dying trees. The earlier the death time of the dying tree was, the higher the isolation rate of *R. quercivora* was, and the rate of the discoloured area of the wood was also higher, and the number of gallery which succeeded in the propagation was also higher. For the above reasons, it was completely proved that *P. quercivorus* was vector of *R. quercivora,* and the mass mortality of Japanese oak trees was caused by mass attack of *P. quercivorus*.

A study on preimaginal development of sweetpotato whitefly *Bemisia tabaci* Gen (Hom.: Aleyrodidae) on melon, cucumber and okra

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Primaginal development of sweetpotato whitefly *Bemisia tabaci* Genn. (Hom., Aleyrodidae) were studies on Melon, Cucumber and Okra under laboratory conditions in temperature 15,25, 30 °C R.H. 60%±5 and photoperiod L:D 14:10 hours. The result showed that total preimaginal development on Melon were 51.51,21.42 and 15.7 days at 15,25 and 30 °C, respectively. On cucumber and Okra this rate was 51.97, 20.99, 13.42 and 66.7, 32.13 and 17.02 days at 15,25 and 30°C, respectively. Statistical calculations showed that there are significant variation among preimaginal development of this pest on melon, cucumber and okra at 15°C at 25 and 30°C , there are significant variation between egg and nymphal stages on okra, melon and cucumber but there aren't any significant variation at pupal stage on these three plants.

Cysteine proteases in latex protect papaya and fig trees from herbivorous insects

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The primary roles of proteins and enzymes often found in plant latex are as yet poorly understood. We found that cysteine proteases in latex of the Papaya tree (*Carica papaya*, Caricaceae) and a wild fig (*Ficus virgata*, Moraceae), are crucial factors in the defences of these plants against lepidopteran larvae such as oligophagous *Samia ricini* (Saturniidae) and two polyphagous pests, *Mamestra brassicae* (Noctuidae) and *Spodoptera litura* (Noctuidae). Leaves of the two plant species showed strong toxicity and growth inhibition against lepidopteran larvae. When the latex was washed off, the leaves lost toxicity. Latexes of both species were rich in cysteine-protease activity. E-64, a cysteine protease-specific inhibitor, completely deprived the leaves of toxicity when painted on the leaf surface. Cysteine proteases, such as papain, ficin, and bromelain, all showed toxicity. The results suggest that plant latex and the proteins in it, cysteine proteases in particular, provide plants with a general defence mechanism against herbivorous insects.

Carrot psyllid *(Trioza apicalis)* host plant ecology – studies of perception, selection and migration.

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The carrot psyllid, *Trioza apicalis* (Homoptera: Psylloidea), is a serious pest on carrots in northern and central Europe. Up to date, monitoring and control methods are inadequate, and the species seems insensitive, if not resistant, to pesticides. Carrot psyllids are univoltine and adults hibernate on conifers. In early summer they appear in carrot fields, where reproduction and egg laying take place. Fairly little is known of the behaviour and biology of this species. In order to suggest alternative, sustainable control and monitoring methods for carrot psyllids, we study their ecology and behaviour. Our goals include identifying host plant odours, and possibly also pheromones, that may be used as attractants or repellents in improved monitoring and push-and-pull strategies. Relating to host plant ecology, we also wish to tease out the factors that control migratory behaviour, host alternation and host choice. Field studies have provided some information about migratory abilities and winter host preferences. In the lab, behavioural assays and extractions of semiochemicals have been unsuccessful so far. To guide electrophysiological efforts, we have studied antennal morphology, revealing a very sparse setup of sensilla. This may suggest that this species is less reliant on olfaction than anticipated.

Symbiotic control of Pierce's disease: the social life of *Alcaligenes xylosoxidans* var. *denitrificans*.

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Alcaligenes xylosoxidans var. *denitrificans* (Axd) is a symbiotic bacterium isolated from the foregut of the sharpshooter insect, *Homalodisca coagulata* (Say) and is often present in the xylem fluid of several host plants. Axd is a candidate to deliver factors to control (symbiotic control) the grapevine strain of *Xylella fastidiosa* (Xf) that causes Pierce's disease because both Axd and Xf occupy the same niche in vector insects and host plants. In our studies, Axd was not found in predators that fed on Axd-containing sharpshooters. Axd did not establish on or within table grapes or in grapevines inoculated artificially with Axd. Axd did not survive in a variety of red and white wines. The bacterium also did not establish in a variety of soil types unless the soil was first sterilized. Axd catabolized a variety of organic compounds but did not produce certain extracellular enzymes considered to be virulence factors. An auxotrophic subpopulation that requires cysteine for growth was isolated. Competition studies using *Pseudomonas* spp., *Escherichia coli* and *Staphylococcus aureus* revealed the influence of environmental conditions on survival and growth of Axd in mixed culture.

Nutritional adaptation of galling insects investigated by globular gall of *Trioza shuiliensis* (Yang) on *Machilus japonica* var *kusanoi* (Hyata)

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Nutritional adaptation of a galling psyllid, *Trioza shuiliensis*, was investigated based on anatomy and component analyses. On the analyses of primary metabolites, protein content in gall tissues is 2 folds than that of leaf tissue while the total non-structural carbohydrate 2-3.6 folds. In contrast, nitrogen content is only half as much as in gall tissues than in the leaf tissues. The secondary metabolite flavonoid has 5 folds content in leaf than in gall, but anthocyanidin only one tenth. By photo-pigment analyses, the content is only one tenth for Chl *a* and *b* in gall than in leaf. PPIX in gall is much higher than in leaf. It suggests that there are some troubles during the transformation from PPIX to MGPP in gall tissues in chlorophyll synthesis. The gall should not have the ability to work normally in photosynthesis as indicated by the analyses of the contents of chlorophylls. Anatomy of gall tissue through paraffin sectioning reveals a circling of vascular bundles on the larval chamber, which suggests the gall should play a function to convey assimilates. High nutritional component contents in the gall should come from other tissues and the gall may imitate a sink to provide galling insects with more nutritional benefit than free-living herbivores.

A survey of arthropod-plant relationships in Mokolodi nature reserve, Gaborone, Botswana

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The semi-arid climatic conditions prevalent in Botswana are amenable to a diversity of arthropods that in turn associate with different plant species. A survey was carried out at the Mokolodi Nature Reserve that is located about 15km south of Gaborone city. The nature reserve harbours a savanna type of vegetation. Arthropods found around the vegetation were mainly of the order Hemiptera. They were found to be associated mainly with members of the tree genus *Acacia*. Observations have shown that, several factors have influenced the abundance and associations of this Order with this particular tree genus. Some of these include, egg-laying, feeding habits and pollination behaviour. Acacias are characteristic of typical savanna vegetation, which attracts arthropods by their leaves, barks and flowers throughout the year.

Conserved regulatory elements in the promoters of two allelochemical-inducible cytochrome P450 genes differentially regulate transcription

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CYP6B4, a cytochrome P450 gene from the tiger swallowtail *Papilio glaucus*, is transcriptionally induced in the midgut by dietary furanocoumarins, plant allelochemicals that can crosslink DNA in their UV-activated form. The *CYP6B4* promoter contains an overlapping EcRE/ARE/XRE-xan element similar to that used for basal and xanthotoxin-inducible expression of the *CYP6B1* promoter from the black swallowtail *Papilio polyxenes*. Transfection of the *CYP6B4* promoter: CAT reporter construct into Sf9 cells demonstrates that the basal and xanthotoxin-inducible expression levels observed reflect the relative expression levels of this gene in the midguts of tiger swallowtail larvae. Transfections of mutant *CYP6B4* promoter constructs into Sf9 cells indicate that the EcRE/ARE/XRE-xan element is necessary for *CYP6B4* induction by xanthotoxin but not for its minimal basal expression. In addition to these elements, the *CYP6B4* and *CYP6B1* promoters also contain putative XRE-AhR elements identical to the aryl hydrocarbon response elements present in mammalian phase I detoxification genes. Transfections of *CYP6B4* and *CYP6B1* promoters containing EcRE/ARE/XRE-Xan and XRE-AhR elements indicate that both are induced significantly by benzo(ɑ)pyrene, an aromatic hydrocarbon widespread in the environment, as well as by xanthotoxin, an allelochemical encountered in their host plants.

Speciation in gall thrips on ***Acacia***

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Australian gall-thrips are a phytophagous insect group that parasitise Australian *Acacia* trees by inducing galls and producing brood within the enclosed structure. The *Klaadothrips rugosus* species complex consists of up to 14 host-related 'races,' and in nearly all cases, each 'race' induces a morphologically discrete gall. Current evidence suggests that these 'races' represent sibling species. Of particular interest are three known instances where each of two different 'races' induces different gall structures on the same host species. Several questions emerge. Is the divergence between each race on the same host at a species level? Does speciation occur by host shifting, cospeciation, sympatric speciation, or a mixture of all these mechanisms? We used data from microsatellite and mitochondrial DNA markers to infer genetic divergence between three pairs of these 'races' that each shares a different *Acacia* host species.

Toxicity of pellitorine, an amide isolated from *Piper tuberculatum* Jacq. (Piperaceae) to the pollinator *Apis mellifera* L. (Hymenoptera: Apidae)

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Pellitorine, an amide isolated from *Piper tuberculatum* (Piperaceae), has been studied as a biopesticide against agricultural pests in Brazil. In order to investigate their impact on non-target organisms, particularly pollinator insects, research were performed to evaluate the acute toxicity action of the compound on larvae and newly emerged adults of honeybee *Apis mellifera* by means of contact and ingestion bioassays. Workers in the larval and adult phase were separated in groups, which received pellitorine in different concentrations. The larvae were maintained in their own original cells, receiving feeding and normal care from the nurses. The adults were confined in wooden cages with screens, receiving artificial diet made up of sugar and water (1:1). The concentrations of 40, 200, 1000, 5000 and 25000 ng a.i./individual were obtained diluting pellitorine in 98% ethanol. LD_{10} values of 39.14, 36.16 and 13.79 ng a.i./insect were determined for larvae, for adults by ingestion and adults by contact, respectively. The honeybee larvae were shown to be highly susceptible to the amide pellitorine. The amide pellitorine was shown to be more active when its absorption took place via dermal contact (by topical application).

Effects of coexistence with a water lily *Trapa natans* on embryonic development, larval growth and oviposition process of a water strider, *Gerris nepalensis*

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"This study tries to clarify the influence of coexistence of a water strider, *Gerris nepalensis* with a water lily, *Trapa natans* as a component of symbiosis. *Materials and Methods* Adult *G. nepalensis* and *T. natans* were collected from a water way in Kochi (33ºN) in August, 2002 and used for rearing experiments under 15h light-8.5h dark, at 25±2°C. Offspring of *G. nepalensis* were also used. Coexistence with the waterlily leaf during larval or adult stage caused shorter preoviposition period (Mann-Whitney U-test, $P<0.05$) and higher number of eggs laid ($P<0.01$) by *G. nepalensis*. Larval period with the waterlily leaf was shorter than that with a mimic leaf made of thin (1mm) sryrene form ($P<0.01$). Eighty percent of eggs laid by females which were reared with the lily leaf in larval and/or adult stage(s) were successfully developed and higher than forty percent without the lily leaf (χ^2-test, $P<0.01$). Co-existence of *G. nepalensis* with *T. natans* seems to make the individual number of *G. nepalensis* increase and lead to higher foraging pressure to the lily leaf beetle, *Galerucella nippnensis*. Symbiosis between *T. natans* and *G. nepalensis* exists via *G. nippnensis*.
"

Resistant mechanism of tea plants, *Camellia sinensis* **O. Kuntze to mulberry scale,** *Pseudaulacaspis pentagona* **Targioni (Hemiptera: Diaspididae)**

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The mulberry scale, *Pseudaulacaspis pentagona* is the most serious pest of tea plants in Japan. The control effect by chemicals against *P. pentagona* was insufficient, since the insect settling in the tea bushes. The use of resistant cultivars against *P. pentagona* is one of the effective control methods. The resistant cultivars such as Sayamakaori and Minamisayaka, inhibited the development and reproduction of *P. pentagona*. The total number of laid eggs and ovarian eggs per female adults on the resistant cultivars was 5 to 10 % of that on the susceptible cultivar, Takachiho. Moreover, there was a significant positive correlation between the egg reproduction per female and the degree of infestation in tea fields. The weight of scale cover in females reared on the resistant cultivars was significantly lighter than that on the susceptible ones, suggesting that nutritional ingestion was insufficient in the former. When the stylet of female adults was severed, the frequency and amount of exuding phloem sap through the severed stylet was significantly lower on the resistant cultivars than that on the susceptible ones. This difference is presumably related to the mechanism of antibiosis against *P. pentagona*.

Effects of experience of *Cassida nebulosa* L. (Coleoptera: Chrysomelidae) on preference for spinach

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A tortoise beetle, *Cassida nebulosa* feeds on chenopodiaceous weeds, *Chenopodium album* and *C. album* var. *centrorubrum* as its main hosts. When the adults were reared on a chenopodiaceous plant, spinach, they feed on spinach as well as *C. album* var. *centrorubrum*. However, the adults reared on *C. album* var. *centrorubrum* did not consume spinach immediately. Methanol extracts of spinach showed a strong feeding-deterrent effect to *C. album* var. *centrorubrum*-reared adults. An active fraction was separated from methanol extracts by solvent partitioning, ODS and silica gel column chromatography. The feeding-deterrent activity on spinach-reared adults was less than that on *C. album* var. *centrorubrum*-reared adults. In addition, when adults were reared on leaves of *C. album* var. *centrorubrum* treated with the active fraction, they accepted spinach as well as *C. album* var. *centrorubrum*. These results indicate that exposure to the deterrent reduce deterrent activity on *C. nebulosa*. Therefore, alternation of activity of the deterrent contained in spinach is responsible for difference of acceptability of spinach by different feeding experience.

Parasitoid communities of herb gall wasps (Hymenoptera:Cynipidae: Aylacini) from Europe

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We compiled literature data and unpublished qualitative and quantitative information on the parasitoid community (Hymenoptera Chalcidoidea) of herb gall wasps (Hymenoptera, Cynipidae, Aylacini) from Europe. This essay is aimed to contribute the knowledge of the composition and diversity of one of the most complex, structured and diverse insect communities associated to insect galls. The host gall community is composed of 40 cynipid species inducing galls on 21 plant genera from four families (Asteraceae, Lamiaceae, Papaveraceae and Rosaceae). The studied associated parasitoid community accounted more than 12.000 individuals from 101 species belonging to six chalcidoid and one figitid families as follows: Eulophidae (9), Eupelmidae (9) , Eurytomidae (24), Ormyridae (10), Pteromalidae (25) and Torymidae (23) (Hym., Chalcidoidea); Figitidae (1) (Hym., Cynipoidea). The community presented a high percentage of monophagous, specific-associated parasitoids (about 56%) whereas the estenophagous and poliphagous components were much lower (about 25% and 20%, respectively). Some of the most species-rich parasitoid communities were from cynipids species inducing cryptic galls, indicating that herb galls wasp diversities could be non dependent of size or structural complexity of the host galls.

Two new species of herb gall wasps of the genera *Iraella* and *Aulacidae* (Hymenoptera: Cynipidae: Aylacini) from Spain, including description of their terminal larvae and galls

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Two new species of herb gall wasps (Hym., Cynipidae, Aylacini) are described from Spain: *Iraella hispanica* sp. n. and *Aulacidea martae* sp. n. *Iraella hispanica* sp. n., was collected in Tarragona and Zaragoza (North Eastern Spain). The new species represent the second described species from the former monotypic *Iraella* genus and is related to *I. luteipes* that induces cryptic galls in stems of the opium poppy. However galls of the new species are formed in common poppies heads (*Papaver spp*, Papaveraceae) and for this feature is also related to species of the genera *Aylax* and *Barbotinia*. Galls of the new species showed high percentages of parasitism by the figitid species *Parnips nigripes* (Cynipoidea, Figitidae). *Aulacidea martae* sp. n. induces cryptic galls on twigs of *Launaea arborescens* (Asteraceae), a plant living in sand dunes from the Natural Park of Cabo de Gata-Nijar (Almeria), a protected desert habitat at South Eastern of Spain. The host plant of the later new species is recorded for the first time as host of a cynipid gall wasp. Adults of the two new species are described and fully illustrated with SEM pictures. Data are also given on the biology of the two new species described, including description of their terminal larvae and galls.

Morphology and biology of terminal-instar larvae of Ormyridae (Hymenoptera, Chalcidoidea)

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Comparative larval morphology of nine ormyrid species (Hym., Chalcidoidea, Ormyridae) from Europe is for the first time presented. The species are representatives of the different life cycles and host plant associations presented for the family in Western Europe: *O. pomaceus*, *O nitidulus*, *O. rufimanus*, *O. diffinis*, *O. wachtly*, *O. capsalis* and *O. papaveris*, parasitoids of oak and herb gall wasps; *O. cupreus*, parasitoid of eurytomids inducing galls on *Ephea* and *O. orientalis*, a parasitoid of tephritid flies in head flowers of *Centaurea* species. The terminal-instar larva of the listed species was prepared for observation under SEM at low vacuum and pictures were taken of the ventral body, head, mouth parts and mandibles. We describe the external ormyrid larval morphology, comparing with the larva of related chalcidoid groups, namely Eurytomidae and Torymidae, as well as we summarize the qualitative variation among the studied larvae. Some diagnostic characters could characterize the family: presence of scarce and short pilosity on the body, rough tegument on parts of head and first segments of body, one toothed mandibles, and a trilobate labrum without deep divisions in the centre.

Bioinsecta - software for insect biology analyses

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The BIOINSECTA is software developed in WindowsÒ platform with the finality of helping researchers of the entomology area to execute analyses related to insect biology study. This software contains pre-established spreadsheets that can be used to evaluation of experiments related to the insect's biology, facilitating this way, the annotation and organization of data to insertion in the program. Trough a simple interface and easy application, the user creates, her/his data bank that can be as stored as impressed. During the typewriting of data to create the archive, the user can visualize in real time, the results from biological parameters that are being calculated by the program with the application of pre-established mathematical models. The calculated parameters can be visualized in a separated screen, and they'll be stored for posterior application or impress, according the user choice. Its creation wait to facilitate the biological calculus and reduce the waste time with its execution, without lose the necessary accuracy. This software has been in phase of analysis for register, but a Demo version will be able to be found at: www.bioinsecta.hpg.com.br

Diversity and biology of gall-inducing Lepidoptera in Costa Rica

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Thus far some 100 species (*ca.* 9% of arthropod-induced galls in Costa Rica) of gall-inducing Lepidoptera belonging to at least 10 families and 8 superfamilies have been found, and are associated with *ca.* 80 species of vascular plants belonging to 20 plant families of dicotyledons. More than half of these lepidopterans are undescribed species of *Mompha* (Coleophoridae), nearly all of which occur on Melastomataceae. Other lepidopterans are: Nepticulidae (1 sp.), Gracillariidae (3), Glyphipterigidae (1), Elachistidae (4), Cosmopterigidae (7), Sesiidae (5) Tortricidae (5), Alucitidae (3), Thyrididae (1). Remaining lepidopterans have not yet been identified. Galling is unknown in butterflies or any other lineage of Macrolepidoptera. Galls were found on stems, leaf veins, petioles, flowers, fruits, and roots. The galls are induced by the larva hatching from an egg laid on the host plant surface and range from simple inconspicuous swelling to complex tissue and organ modification. Many inducers spend their entire larval and pupal stages inside their galls, while others pupate away from the gall. Excrement is packed in the gall chamber or ejected through an orifice. As in other gall formers, gall-inducing Lepidoptera are highly host specific and some could be potential biological control agents for invasive weeds.

Role of PGR-brassinosteroids on the development of a plant-parasitic nematode

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Brassinosteroids, a recently explored group of plant growth regulators, controls a spectrum of physiological responses in a variety of test systems. Their effects on some insects and animals have been well documented in literature but their effect on nematodes has not been studied much. Nematodes have a very intimate association with their host plant. Any change occurring in the host plant is bound to effect the development and physiology of the associated nematode. Keeping this in mind, the present piece of work envisages the role of isolated (mixture of brassinosteroids) and synthetic (28-homobrassinolide) brassinosteroids on the development of a plant-parasitic nematode, *Meloidogyne incognita*. The egg masses of *M. incognita* were exposed, *in vitro*, to six different concentrations (10^{-5}M to 10^{-10}M) of isolated brassinosteroid and 28-homobrassinolide for 7 days. Observations recorded, revealed an enhanced percentage of hatching in treated egg masses when compared with control. Hatched/hatching juveniles were further allowed to develop on plants for 45 days. Brassinosteroids treated juveniles resulted into more gall numbers and larger sizes of galls as compared to untreated ones.

Amount of green leaf volatiles emitted from *Arabidopsis* plants affects their attractiveness to parasitic wasps of cabbage butterfly larvae

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When infested with cabbage butterfly (*Pieris rapae*) larvae, cruciferous plants emit volatiles that attract a specialist parasitic wasp, *Cotesia glomerata*. This wasp is also attracted to volatiles emitted from mechanically damaged crucifers. Because green plants, such as crucifers, emit six-carbon (C_6-) volatiles through the lipoxygenase pathway in response to mechanical damage, it is likely that some C_6-volatiles are involved in attracting *C. glomerata* to host-infested cruciferous plants. To test the effects of C_6-volatiles on the attraction of *C. glomerata*, Arabidopsis (Cruciferae) seeds treated with mutagens were screened in order to identify mutants in which production of C_6-aldehydes was suppressed (or increased). We obtained a mutant that produced lower levels of C_6-aldehydes than wild-type plants, and this mutant was designated aldehyde less (*all*) *84*. *All 84* also produced lower amounts of other C_6-volatiles when compared to wild-type plants. Laboratory bioassays showed that *C. glomerata* were attracted to *all 84* plants infested by *P. rapae* larvae over uninfested ones. However, when comparing *P. rapae*-damaged *all 84* plants to *P. rapae*-damaged wild-type plants, the wasps preferred the wild-type plants. These data suggest that the amount of C_6-volatiles emitted from *P. rapae*-infested crucifer plants in part determine the degree of attractiveness to *C. glomerata*. Herein, we also discuss the possible effects of a modification in the lipoxygenase pathway of *Arabidopsis* on wasp attraction.

Influence of feeding of *Frankliniella occidentalis* Pergande (Thysanoptera: Thripidae) on the polyphenolic complex in the leaves

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Production of polyphenolic complex compounds in plants contributes to protecting them against herbivores and often determines their forage nutritional value and the insect preference for them. The thrip *Frankliniella occidentalis* is one of the most important insect pests in vegetable crops in greenhouses in the island of Crete (Greece). We recently showed that its preference for several vegetables is variable, ranging from highest levels for eggplants to minimum ones for tomato plants. To test whether this differential preference was associated with the production of polyphenolic compounds in the leaves, we compared the profile of polyphenols, in control (undamaged) and damaged leaves (evoked by *F. occidentalis* attack) of eggplant and tomatoes, by using reverse phase HPLC. We showed that feeding of *F. occidentalis* causes alterations in the relevant quantity of several phenolic acids (chlorogenic acids) and flavonic glucosides (rutins), which are believed to play an important role in plant defence against insect pests. These alterations were different in tomato compare to eggplant leaves, indicating that changes in the production of polyphenolic complex compounds might be associated with the differential preference of *F. occidentalis* for these vegetable hosts.

Canopy management and variety morphology influence in the European grapevine moth (*Lobesia botrana* Den. y Schiff.)

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The influence in the European grapevine moth attack of the canopy management in some of the most important Spanish vine varieties can be modified by different morphology features, in special the cluster structure. The work was carried out during two years, 2000 and 2001, in different plots in Castilla y León (Spain). Four varieties: Tempranillo, Garnacha, Viura and Verdejo, were studied under two different training systems: one trellis-training system with vertical shoot position and the second one, head system without shoot position. In general, there are two generations in the Spanish North Meseta, with two different kind of damage. The goal of this study was to evaluate how can influence canopy management, and other features like cultivar susceptibility of clusters or grapevine phenological stages, on the clusters damages.

Chemical characteristics in the vineyard wood and its possible relationship with *Xylotrechus arvicola* attacks (Olivier, 1795) (Coleoptera: Cerambycidae)

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Xylotrechus arvicola (Olivier, 1795) (Coleoptera: Cerambycidae) is a cerambycid which attacks vineyards. The larvae tunnel into the stem in all directions. This is causing economic damages to grapevines in some regions of Spain: Castilla – La Mancha, Castilla y León and La Rioja, because there is a decrease of vascular capacity. The target of this work is to establish the possible relation between the percentage of infested vines and the variety of vines. For this, we have chosen a vineyard in the D.O Bierzo (León). In this, we have selected three varieties: Tempranillo, Sauvignon Blanc and Mencía. During the winter 2003 - 2004, we have prospected the external symptoms of this insect: tunnels made for larvae and round exit holes made for adults. Also, we have analyzed in laboratory some chemical characteristics of wood: holocellulose, lignin and extracts, in some vines of these varieties. The results showed that Mencia is the variety less attacked in these years and Tempranillo is the most attacked. We have made an ANOVA. There are significant differences in holocelulosa, lignin and extracts between the less and more attacked varieties by *Xylotrechus arvicola*. The highest percentage of holocellulose in the most attacked variety offers to the insect a best quality of food. The highest percentage of extracts in Mencia variety could mean that they act as inhibit substances of larval development or as phytoalexin.

Novel activity from *Piper* (Piperaceae) plant secondary compounds against insecticide-resistant and susceptible insect pests of the home and garden

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Extracts from two pepper species, *Piper nigrum* L. and *P. tuberculatum* Jacq., were tested against insect pests of vegetables and ornamentals in laboratory trials. Based on *P. nigrum* 24 hr LC_{50} values, the larvae of introduced pine sawfly (0.01%); eastern tent caterpillar (0.02%); European pine sawfly (0.05%) and forest tent caterpillar (0.05%) were more sensitive than viburnum leaf beetle (0.1%); gypsy moth (0.28%); Colorado potato beetle (0.5%) and spruce budworm (1%). 24 hr LC_{50} values for *P. tuberculatum* extract showed that both Pyrethroid-resistant house flies and multiple-insecticide-resistant Colorado potato beetles were as sensitive or only two-fold less sensitive than susceptible strains of each species. Preliminary experiments using synaptosomes found that piperamides do affect neuronal Ca^{2+} influx and glutamate release. This evidence coupled with the demonstrated inhibitory effect of piperamides on insect microsomal activity indicates a unique dual activity for these botanical insecticides. *Piper* botanicals therefore have the potential to provide an alternative control product against insect pests of economic importance for organic and conventional horticulturalists, even with populations which have developed resistance to widely used synthetic products.

Arabidopsis thaliana **as a model host plant to investigate the effect of a cyanogenic glucoside on host plant-** *Plutella xylostella*- *Cotesia plutellae* **interactions**

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The presence of cyanogenic glycosides in plant material has been shown to confer resistance to some herbivorous pests. Cyanogenic glycosides have not been found naturally in members of the Brassicaceae. Therefore specialist Brassicaceae pests, such as *Plutella xylostella,* are unlikely, in evolutionary terms, to have encountered cyanogenic glycosides recently as natural constituents in their feed and so been exposed to any direct selection pressure to evolve detoxification processes specific to cyanogenic glycosides. Because of this, the effect of cyanogenic glycosides on previously unexposed pests could have very interesting applications for potential pest control strategies. The recent availability of lines of the Brassicaceae *A. thaliana* that have been transformed to produce a cyanogenic glycoside, dhurrin, make it possible investigate such scenarios. The aim of these studies was to use the transformed *A. thaliana* lines to investigate the effect of dhurrin, on the herbivore pest *P. xylostella* and its specialist parasitoid *C. plutellae.* Dhurrin expression did not affect *P. xylostella* in terms of growth, development, fecundity or preference. *C. plutellae* development time was extended by dhurrin, and at lower concentrations, the resulting parasitoid was larger. Dhurrin also disrupted the processes of olfactory choice in host searching by the parasitoids. These studies suggest that dhurrin is not advantageous to protection of the host plant from *P. xylostella.*

Oviposition site selection and phenotypic plasticity in the number of larval instars of a lichenivorous *E. depressum*

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Although one might intuitively expect a tight correspondence with female host preference and larval performance, several studies have reported relatively poor preference-performance correlations in herbivorous insects. I compared the host preference and subsequent larval performance of lichenivorous *Eilema depressum* on four common, epiphytic lichens. Females did not show clear host preference towards any of the host lichens. However, *Melanelia exasperata*, on which larvae grew faster and bigger than on other lichens, resulted in the best performance measures of larvae. Moreover, the number of larval instars varied between 6 and 13. Over-wintering larvae had considerably more instars than larvae, which developed without over-wintering to adult stage. Host lichen also had a weak impact on the number of instars. I discuss the importance of larval responses to nutritionally varying hosts in the absence of host preference of ovipositing females and the significance of phenotypic plasticity in the instar number of a lichenivorous species.

Micromorphology of the mouthparts of specialist eucalypt browsing cockchafers *Heteronyx* spp.: an insight into adaptation to a novel adult host plant

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The adaptation of morphological structures facilitating host specialization is a key factor in insect-host associations. Endemic phytophagous Melolonthine cockchafers are serious pests during the establishment of exotic *Eucalyptus nitens* plantations in Tasmania. The mouthparts and associated sensory structures of two species, *Heteronyx crinitus* Blackburn and *H. dimidiata* (Erichson) were examined by ESEM for specialization. *H. crinitus* displayed highly developed mandibles capable of excising tough foliage of its *Eucalyptus* host. Defences of the *Eucalyptus* host such as leaf toughness seem insufficient to deter scarab browsing. The modified environs of production forestry exposes *Eucalypt* seedlings to attack by native insects which evolved with other hosts in the near vicinity.

Application of multivariate analysis to electrical penetration graphs using manual and automate waveform recognization from the planthopper *Peregrinus maidis* feeding on susceptible and resistant maize

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The stylet penetration activities of *Peregrinus maidis* (Ashmead) (Homoptera: Delphacidae), vector of *Maize mosaic virus* (MMV) and *Maize stripe virus* (MStV) were studied by electrical penetration graphs (DC-EPG) on resistant and susceptible maize inbred lines. Multivariate analyses were applied to two sets of data obtained by manual and automate methods of waveform recognization from digital EPG recordings. For both methods, the variables, which mostly explained the resistance-susceptibility status of the maize lines, appeared to be related to the passive ingestion in phloem vessel but also to the stylet pathway and in a less extent to active ingestion in xylem and to non-penetration. In spite of a great variability within each genotype, three groups of individuals could be distinguished separating genotypes assumed as susceptible, slightly resistant and resistant to the virus transmission. These results are discussed in relation to the possible mechanism of maize resistance to *P. maidis* and the usefulness of automate waveform recognition with the software "EPG soft" for plant breeding.

Feeding behaviour of the planthopper *Peregrinus maidis* on maize analysed by digital signal processing of EPG and MET structure of stylet pathway

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Peregrinus maidis (Asmead)(Hemiptera, Delphacidae) is the only known vector of *Maize mosaic virus* and of *Maize stripe virus*, both circulative and propagative viruses. In order to better understand the feeding behaviour of *P. maidis* the electrical penetration graph technique was used (DC-EPG), according to this tool five different waveforms were characterised. Based on the correlation between waveforms and the fine structure of stylet pathways observed by transmission electron microscopy, active ingestion was restricted to xylem vessels, and passive ingestion and watery salivation occurred intermittently in sieve tubes. Spectral and time-frequencies analysis of digital data obtained with the "EPG-Soft" software allowed us to better distinguish ingestion and salivation activities in sieve tubes. Ingestion and watery salivation are essential phases of the feeding behaviour of piercing sucking insects, respectively correlated with acquisition and inoculation of circulative viruses.

Three new host plants of the *Brevipalpus*-borne virus found in United States

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Mites within the genus *Brevipalpus* (Acari: Tenuipalpidae) have been incriminated with the transmission of a series of new viruses in South America. During a survey for mites and viruses in ornamental plants in Florida, the following plant species had ringspot symptoms in association with *Brevipalpus* mite infestations: *Pittosporum tobira* (Thunberg) Aiton (Pittosporaceae), *Hedera canariensis* Willdenow (Araliaceae) and *Trachelospermum asiaticum* Nakay (Apocynaceae). Transmission electron microscopy of plant tissues within the lesion areas of each of the three species showed rod-like particles within the cell nucleus and one prominent nuclear viroplasm, as well as particles accumulated in the cytoplasm adjacent to the nucleus. These cytopathological alterations in the cells are typically linked with the 'nuclear type' of viruses associated with *Brevipalpus* mite vectors. So far, the virus associated with *Hedera* has been experimentally transmitted by *Brevipalpus* mites.

A rapid method in laboratory to screen resistance of potato plants against *Myzus persicae*

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In the objective to develop potato resistance against aphids, we propose a rapid screening method with *Myzus persicae* (Sulzer) in laboratory. Our study focussed on two objectives, i.e. to optimize the duration of the whole procedure and to decrease the frequency of measurements. In a first experiment, no significant differences were observed between intrinsic rate of natural increase (r_m) values of adult aphids reared throughout their entire life and adults reared only during a period equivalent to their pre-reproductive period. In a second experiment, four groups of aphids were distinguished according to the sampling frequency, i.e. those whose biological parameters were evaluated every single, second, third and fourth day. Except for the fourth day experiment, the r_m values estimated every single, second and third day were not significantly different for all potato lines. Thus screening efforts in laboratory can be largely optimized by checking adult aphids only during a period equivalent to their pre-reproductive period and assessing *M. persicae* populations every third day. Our method is reliable and adapted to screen a large number of potato plants against *M. persicae* as it leads to an average 70% reduction in the time required for the whole experimental process. Our laboratory screening method must be complemented with a field-screening program.

Woolly apple aphid electrical penetration graph - 'A' waveform

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Molecular biological analysis on the induced resistance to pathogens in rice plants infested by *Sogatella furcifera*: expression of defense-related genes and induction of pathogenesis-related (PR) proteins

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We found that infestation of white-backed planthopper, *Sogatella furcifera*, induced significant resistance to rice blast fungus-infection in the rice plants; the number of lesions formed on rice leaves by infection of blast fungus, *Magnaporthe grisea*, was quite few in the plants which had been exposed to the planthoppers compared with control plants. When exposure was restricted to stem, the number of lesions in the exposed plants was also smaller than that of control plants. The level of incidence by blast fungus-infection after infestation by male planthoppers was similar to that by female planthoppers. Moreover, we found that rice plant which had been infested by the planthoppers acquired resistance to the bacterial blight caused by *Xanthomonas oryzae pv. oryzae*. To study the mechanism of induced resistance, we analysed expression of defense-related genes, accumulation of pathogenesis-related (PR) proteins in the rice plants which were infested by the planthoppers. Interestingly, we found that expression of some defense-related genes and accumulation of PR-1a, PR-2 and PR-5 proteins were considerably increased. These results suggest that the induced resistance by the planthopper mimics the acquired resistance to pathogen-infection in plants which has been extensively studied.

Host selection by pine processionary moth *Thaumetopoea pityocampa* females

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Thaumetopoea pityocampa (Den. & Schiff.) (Lep.: Thaumetopoeidae) is one of the most destructive defoliators of natural and artificially established *Pinus* spp in the Mediterranean region. Infestation rates differ dramatically among species and tree individuals, suggesting female choice of hosts during egg-laying. We have identified two main factors affecting the host selection process: (i) Tree silhouette, suggested as a dominant factor in the literature. In a rearing room we offered to moths several *Pinus nigra* trees, a preferred species, of different sizes. A significant positive correlation was found between the number of egg masses laid on each tree and both tree height and silhouette. (ii) Volatiles of host plant may play an important role in habitat selection. We investigated the olfactory host recognition mechanisms of females by the gas chromatographic-electroantennographic technique (GC-EAD). We tested natural volatile blends and a synthetic mixture of monoterpenes present in pine needles. Repeatable responses were among others to minor compounds like *E*-ocimene, -terpinol and neodihydrocarveol. In addition, the identification of suitable needles through contact sensilla on tarsi is evaluated as an additional input for the host choice by females.

The suitability of honeydew as a food source for aphid antagonists

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Honeydew is a carbohydrate-rich excretion produced by sap-feeding insects. Honeydew can be particularly crucial for survival and fecundity of aphid parasitoids, as well as for aphid predators. The suitability of honeydew as a food source can vary largely, depending on the plant and sap-feeder species. Besides different sugars and nitrogen-rich compounds, honeydew may contain secondary plant compounds. Here we address the nutritional suitability of honeydew from two aphid species, *Myzus persicae* and *Brevicoryne brassicae*, feeding on *Brassicae nigra* (secondary plant compound sinigrin, glucosinulate). The two honeydew types were fed to *Chrysoperla carnea* larvae and *Aphidius colemani* adults, to determine their effect on the survival of these two antagonists. In addition, *C. carnea* was fed with the aphids directly. Secondary plant metabolites present in the honeydew and present in the insects were analysed by HPLC in order to trace their pathways. The results show significant differences in the amount of secondary plant compounds in the honeydew from the two aphid species as well as temporal differences relative to the age of the aphid colony. These patterns correspond with the differences in survival of the natural enemies in the longevity experiments. Tritrophic effects in this plant-herbivore-antagonist system are discussed.

Interactions between Caryophyllaceae species and their pollinating fruit predators

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The larvae of all noctuid *Hadena* species and of *Sideridis rivularis* feed exclusively on Caryophyllaceae species, mostly on the fruits. As shown for *Hadena bicruris* and its important dioecious host plant *Silene latifolia*, the moths reproduce within female flowers they pollinate. The moths are very specific by the choice of plants for egg deposition. Also, comparisons of pollen spectra, prepared from different museum specimens and species indicated in some cases a narrow nectar-plant spectrum of the moths. In case of *H. bicruris* and *S. latifolia* it was demonstrated that the nocturnal flower scent attracts the moths, however, nothing was known about the role of single scent compounds. We identified about 60 different floral scent volatiles, and revealed a high variability in the scent of *S. latifolia*. Sixteen electrophysiologically active compounds were identified using the GC-EAD method. Wind tunnel bioassays indicated that some of the EAD active compounds are very attractive for *H. bicruris*. Differences in the chemical composition of total-flower volatiles and pollen volatiles could be important for *H. bicruris* to distinguish between male and female flowers, because eggs are deposited only in female flowers.

Orientation of larvae of gram podborer, *Helicoverpa armigera* (Hübner) (Lepidoptera: Noctuidae), to hexane extracts of susceptible and resistant varieties of pigeonpea, *Cajanus cajan*

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Orientation response of neonate larvae of the gram podborer, *Helicoverpa armigera* (Hübner) to crude hexane extracts of leaves, buds, flowers, and pods of susceptible (ICP 11965) and resistant (ICP 11957) cultivars of pigeonpea was studied in the olfactometer under choice conditions. Extracts from the susceptible variety presented at one end against an opposite blank elicited significantly greater attraction of the larvae towards all the test samples. Compared to the blank end, there was greater orientation towards extracts from leaves, buds, flowers, and pods of the resistant variety. In both the cases attraction was highest for flower extract and lowest for leaf extract. In two-choice tests, the orientation response towards extracts of buds, flowers, and pods of the susceptible variety was significantly higher compared to extracts of the same parts from the resistant variety, but the response for the leaf extracts was similar for both varieties. The orientation preference of larvae for extracts from pods, buds, and flowers of the susceptible variety was respectively 35 %, 41 % and 56 %. This indicates presence of hexane-extractable semiochemicals in the reproductive parts of the plants, contributing towards the greater arrival of podborer larvae, and hence possibly one of the mechanisms of relative susceptibility of ICP 11965.

Mother doesn't always know best – oviposition preferences of *Delia radicum*

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For insect larvae with limited mobility, the choice of oviposition host by the adult female largely determines the plants on which the offspring feed. In greenhouse trials, laboratory-reared cabbage root maggot *Delia radicum* L. female flies oviposited significantly greater numbers of eggs near yellow mustard *Sinapis alba* roots than near oilseed rape *Brassica napus* plants. This ovipositional preference was maintained in trials comparing each of four *S. alba* lines varying in glucosinolate and erucic acid levels with the control *B. napus* cultivar. Although previous research has shown that females prefer larger plant crowns on which to oviposit, in the current study *B. napus* plant crowns were larger than those of *S. alba*. Despite this ovipositional preference, roots of *S. alba* consistently supported fewer *D. radicum* maggots than did those of *B. napus*, a condition frequently found in the field. The possible reasons behind this anomaly are discussed. In this case oviposition preference is not a reliable measure of host plant resistance towards cabbage root maggots. *S. alba* appears to have components detrimental to the survival of *D. radicum* larvae.

Feeding behaviour of *Myzus persicae* on potato plants infected by potato leafroll virus

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Potato leafroll virus (PLRV) is highly dependent upon *Mysus persicae* for dispersal and transmission. Indirect interactions between PLRV and *M. persicae* occur through their mutual host plant *Solanum tuberosum* L. Any change in virus-infected host plants attracting and benefiting the insect vector would increase the probability of virus dispersion. In order to get a better idea of vector-host-virus interactions we studied the effect of PLRV infection on the feeding behaviour of *M. persicae*. We investigated this effect using the electrical penetration graph (EPG) technique, on a potato cultivar that shows a certain degree of resistance to the vector. This resistance avoided phloem sap ingestion (needed for PLRV acquisition) but did not avoid phloem salivation (needed for PLRV inoculation). The feeding behaviour was monitored in control (healthy) and virus-infected potato plants in three different stages; 1) before inoculation with PLRV, 2) 21 days after inoculation, when virus infection was confirmed by ELISA, and 3) when symptoms of the disease were expressed. An increased susceptibility, allowing phloem ingestion (PLRV acquisition) would be beneficial for the PLRV spread but not for the farmer.

Sequence analyses of two plant rhabdovirus genomes and identification of virus-binding insect vector proteins

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Insect transmission of rhabdoviruses is highly specific; a given rhabdovirus is transmitted only by one or a few closely related insect species. *Maize mosaic virus* (MMV) is transmitted by the planthopper *Peregrinus maidis*, whereas Maize fine streak virus (MFSV) is transmitted by the leafhopper *Graminella nigrifrons*. The goals of this study are to sequence the genomes of MMV and MFSV and to identify and characterize factors essential to vector specificity. The MFSV genome has seven open reading frames (ORFs) on the antigenomic strand in the order 3'-N-P-3-4-M-G-L-5', which is one ORF more than MMV having six ORFs in the order 3'-N-P-3-M-G-L-5'. Gene junctions with consensus sequences separate each ORF. *In planta* subcellular localization of fluorescent protein fusions was used to confirm the annotation of MMV and MFSV genes. MMV and MFSV-binding proteins of vector and non-vector insects were determined by virus overlay assays of one-dimensional Western blots. A 90 kDa *P. maidis* protein that bound to MMV and MFSV and a 65 kDa *P. maidis* protein that bound to MMV but not MFSV are being purified by two-dimensional protein electrophoresis and sequenced using tandem mass spectrometry.

The same sort of gall induced by different *Asphondylia* species (Diptera: Cecidomyiidae) on the same host organ and species

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Many gall midges (Diptera: Cecidomyiidae) are monophagous or oligophagous and gall morph is specific to gall-midge species. Therefore, the gall is regarded as an extended phenotype of respective gall midges, except for some polyphagous species. In addition, there are many examples in which different sorts of gall are induced by congeners on the same host organ and species. However, we found three examples in which different *Asphondylia* congeners induce the same sort of gall on the same host organ and species: the chili-pod gall midge in the Mediterranean region (= *Asphondylia gennadii*) and Southeast Asia (*Asphondylia* sp.); the soybean-pod gall midge in Japan (= *Asphondylia yushimai*) and Indonesia (*Asphondylia* sp.); the *Ampelopsis*-fruit gall midge on the Nansei Islands, Japan (*Asphondylia* sp.) and in other areas of Japan than the Nansei Islands (= *Asphondylia baca*). We examined morphological and genetic differences between the congeners that induce the same sort of gall on the same host plant. Their ecological features including host-plant utilization and life-history pattern were also surveyed. Then, we concluded that the congeners on the same host plant were phylogenetically apart from one another. This implies interesting processes in the establishment of gall midge and host plant association.

Identification of sieve elements as the primary site of whitefly resistance in resistant alfalfa genotypes

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Multiple techniques indicate that the mechanism of resistance in alfalfa genotypes previously identified as resistant to silverleaf whitefly, *Bemisia argentifolii* Bellows & Perring, reside in phloem sieve elements. Whiteflies experience very high mortality in the first instar on resistant genotypes compared to susceptible genotypes, but are equally successful in reaching vascular bundles and penetrating phloem sieve elements with their stylets on both genotypes. Phloem sap ingestion commences equally on both resistant and susceptible genotypes, but bouts of ingestion behaviour, as monitored by EPGs, are shorter on resistant genotypes than on susceptible genotypes. Conversely, bouts of a non-ingestion EPG waveform (presumably salivation) are longer on resistant than on susceptible genotypes. Honeydew production per unit time of ingestion is greatly reduced on resistant compared to susceptible genotypes. The combined effects of reduced time in ingestion behaviour and reduced amounts of honeydew produced per unit time of ingestion behaviour result in a drastic reduction of total honeydew production on resistant genotypes. Honeydew production on resistant genotypes ops to near zero levels about 2-3 days after hatching, and the nymphs fail to molt to the second instar.

Attraction of *Hyalesthes obsoletus* **to the Chaste Tree,** *Vitex agnus-castus*

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The planthopper, *Hyalesthes obsoletus (Ho)*, is a known vector of phytoplasmas (the causative agent of "yellows" diseases of grapevines) in Europe and Israel; however, grape is a poor host plant. Therefore, we surveyed native plants in northern Israel to find preferred host plant(s). The chaste tree, *Vitex agnus-castus* L (*Vac*), was found to harbour large populations of adult *Ho*. We planted a belt (alternating groups of 5 plants each) consisting of known *Ho* hosts: *Vac*, *Myrtus communis* and *Convolvulus* sp. We monitored for the presence of *Ho* on these plants, and also at various distances from the belt into the vineyard, by vacuuming. Vacuuming *Ho* from the belt to the centre of the vineyard showed that the number of specimens appeared to decline as the distance from the belt increased. Behavioural studies using a Y-tube olfactometer showed that *Ho* preferred *Vac* over *Convolvulus* sp. or grapevines. Antennal electrophysiological response towards leaf volatiles collected from *Vac* was significantly higher than to grapes. These results suggest that *Vac* is attractive to *Ho* and indicate that host specific volatiles of *Vac* play a role in this phenomenon. Implementation of *Vac* as a trap plant in the control of "yellows" disease is discussed.

Electroantennogram or single cell recordings, what is the best?

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In identifying the volatiles that insects use to locate suitable host plants, electrophysiological recordings of olfactory responses to plant volatiles may give important information. However, divergent results may be obtained with different recording techniques. To illustrate these differences, the results of a previous investigation using single cell recordings linked to a gas chromatograph (SCR-GC) are compared with the results obtained with an electroantennogram linked to a gas chromatograph (EAG-GC). Testing insects of the same species (*Hylobius abietis*) for the same test sample, 30 potent volatile compounds were identified by SCR-GC and 18 by EAG-GC. Of the 34 different compounds, 14 were identified by both techniques. Furthermore, when the same compound elicited detectable responses by both techniques, the response strength was usually not the same relative to the strongest response recorded by each technique. This shows that both EAG-GC and SCR-GC are important techniques in the identification of potent plant volatiles for insects. However, by using SCR-GC more information was obtained, information that can be crucial for understanding the insect-plant relationship.

Defense and regrowth in a willow hybrid population under a nutrient gradient

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The relationship between plant regrowth and defense against herbivores under different nutrient regimes was investigated. We performed an experiment with artificial browsing on two willow species *Salix viminalis* and *S. dasyclados* and six of their F2 hybrids, under three different nutrient regimes (from low to high). We found variation in production of secondary metabolites but no variation in tolerance (biomass of browsed plants/biomass of non browsed plants). In all genotypes the plants in lower soil fertility invested more in root biomass. This may be a mechanism by which plants tolerate herbivory. Our results further suggest that, despite genotype differences in herbivore defense, these are not related to the different growth strategies, rather, the levels of secondary metabolites are related to nutrient levels. We found large effects of genotype on naturally-occurring leaf-hopper damage. However, this does not seem to be related to the levels of secondary metabolites. This in combination with the lack of response in the bioassay with the white marked tussock moth (*Orgyia leucostigma*), shows that the level of insect damage may be a more complicated story than just a matter of how much a plant allocates resources to production of defensive compounds.

Colonization and survival of *Cameraria ohridella* (Lepidoptera, Gracillariidae) on different species of the genus *Aesculus*

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Cameraria ohridella is a small leaf-mining moth which was initially recorded in Europe in 1985 (Deschka & Dimic 1986) and has rapidly colonized almost all its area (Sefrova & Lastuvka 2001). The sudden appearance of *C. ohridella* in Europe has been an unexplained issue so far. One of the likely hypotheses is that it has been brought with plant material from a distant region. The purpose of the presented research is to determine the supposed origin region of *C. ohridella* by observations of moth preferences for host plant. The tests concerning colonization and survival showed that *C. ohridella* went full development on four out of six *Aesculus* species examined and on five hybrids. The highest level of larvae infestation and survival was noted on Asian species: *Aesculus turbinata* and *A. wilsonii*, and on a European species *A. hippocastanum*. The other colonized plant was a North American species *A. glabra*, however the survival of larvae was significantly lower. Two other species from that area, *A. octana* and *A. parviflora*, were not infested.

Life history, gall anatomy and galling position of two urn-shaped cecidomyiid galls on *Machilus* spp. (Lauraceae) in Taiwan

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Cecidomyiid galls represent the most abundant and complicated component of the gall fauna in Taiwan. They comprise about 40% of the galls found in Taiwan but their taxanomy and biology are little known. We studied the life history, gall anatomy and galling position of urn-shaped cecidomyiid *Dephnephila* galls which are commonly found in central Taiwan on two different hosts, *Machilus zuihoensis* and *M. mushaensis* (Lauraceae). Both insects are univoltine. The time of oviposition is influenced by the host phenology in spring and a retardation in development occurred after the hatched first instar larvae mined into the leaf blade until November. There is no correlation between the gall size and the leaf blade size. However, the gall size is influenced by the factors such as galling position, density of galls per leaf and parasitism. Highly differentiated gall tissues can be identified. Fungal mycelia are found on the inner gall surface and they are identified as ambrosia galls. Morphological differences were found for both the insects and their gall which suggest that these are two different species. The morphological consistency of these two similar but distinct galls also provides useful information for resolving the difficulty in identifying *Machilus* species.

Study on host plant selectivity of the leafminer, *Liriomyza sativae*

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The selectivity of six plants species by *Liriomyza sativae* was evaluated with the number of larvae, degree of damage and cucurbitacin B content. The results showed: (1) The Mean number of larvae per cm^2 being 0.183 0.116 0.102 0.099 0.043 and 0.0 respectively for *Vigna sesquipedalis*, *Cucumis melo* var. *conomon* f. *albus*, *Benincasa hispida* var. *chieh-qua*, *Luffa cylinical*, *Cucumis sativus* and *Monordica charantia*. The average degree of larval damage was 23.42%, 13.47%, 8.78%, 5.83%, and 1.63% on *L. cylinical*, *V. sesquipedalis*, *B. hispida* var. *chieh-qua*, *C. melo* var. *conomon* f. *albus* and *C. sativus*, respectively. (2) The levels of cucurbitacin B of the six species were analysed using HPLC. There was a significant negative correlation between the number of larvae, the degree of damage and cucurbitacin B content. Results demonstrate that cucurbitacin B is one of an important factors influencing host selectivity of *L. sativae*.

Annual cycle of the African rice gall midge in southwestern Burkina Faso: host-plant relationships.

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The African rice gall midge, *Orseolia oryzivora*, is a major pest in Burkina Faso. The present study was carried out during 4 consecutive crop seasons from 1999 to 2001 in three rice irrigated schemes in the southern part of the country. It aimed at monitoring the pest damage and its immature stage populations through observations on growing rice in farmer fields, on rice ratoons after harvest and on wild rice around rice fields. The pest survival in absence of rice was due to rice ratoons and wild rice, *Oryza longistaminata*. However, rice crop was the preferred host plant as over 15 % of galls were observed during wet seasons while damages and larval populations were very low in dry seasons. These results are important steps for the development of IPM for the pest control with regard to its host plant relationships.

"Role of pseudomonad induced resistance in tomato *Lycopersicon esculentum* against leafminer, *Liriomyza trifolii*"

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Tomato is an important vegetable crop of India and it is frequently infested by the leafminer, *Liriomyza trifolii* resulting in yield loss. Insecticides offer less impact in its management and genetic resistance iS minimum in commercial cultivars. Induced resistance is found to be a source of pest reduction in crops like tomato. Hence, an attempt was made to induce the resistance in tomato using biological and chemical inducers. *Pseudomonas fluorescens* (strain Pf1) was used to treat the seeds, seedlings and was applied to soil and foliage. The incidence of *L. trifolii* was significantly reduced on plant basis (29.7%) and leaflet basis (19.9%), when compared to check (53.7 and 31.9% respectively). The induced plants showed non-preference for feeding and egg laying to female flies. Antibiosis was observed in larval growth and development. Egg to adult period was increased by 3.2d. The larval parasitism by *Chrysaonotomyia rexia* was 34.7% more on the induced plants. Leafminer infestation resulted in enhanced expression of the pathogenesis related (PR) proteins and defense enzymes *viz.,* peroxidase, polyphenol oxidase, phenylalanine ammonia lyase, 1-3 glucanase, proteinase inhibitors and chitinase. The defense chemicals *viz.,* total phenols, tannins and acylsugars were also high in the induced plants. Compatibility of induced resistance was also noticed with the neem products under field condition.

Quantification of silk trails made by lepidopteran larvae.

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Lepidopteran silk is made up of fibrous proteins extruded by a spinneret located near the mouthparts. The silk produced by lepidopteran larvae is known to play a role in feeding, survival, shelter and dispersal. It is also suspected that silk plays an important role in the adherence of larvae to substrates. Most studies have described the function of lepidopteran silking behaviour in relation to shelter building and construction of pupal cocoons. Our aim was to investigate the role of Lepidopteran silk in foraging behaviours such as crawling and feeding using the larvae of *Helicoverpa armigera* Hübner (Lepidoptera: Noctuidae), a major lepidopteran pest of crops world wide. Given that silk is practically invisible to the naked eye we developed a method for visualising silk trails in early instars using a fluorescent stain and fluorescence microscopy. This was combined with timelapse digital video-recording to correlate larval behaviour and rate of movement with the type and density of silk laid down over the animal's path. Distinctive silking patterns occur in association with larval foraging. Studies were conducted on whole plant material as well as a variety of artificial surfaces, leading to the development of a suitable model substrate.

Begomovirus Transription in *Bemisia tabaci*

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Plant pathogenic begomoviruses have a complex association with the whitefly, their insect vector, and aspects concerning viral genetic activity (genome replication and gene transcription) within their insect vector remain highly controversial. Selected gene transcripts of Tomato mottle (ToMoV) and Tomato yellow leaf curl (TYLCV) begomoviruses were quantified in whiteflies after feeding on virus infected tomato plants and after subsequent transfer to cotton (a non-host for the selected begomoviruses). Real time rt-PCR (RT rt-PCR) was performed using specific primers for three ToMoV genes: AV1, BC1, BV1, and three TYLCV genes: V1, V2, and C3. The ToMoV gene transcripts were rapidly lost in whiteflies transferred from tomato to cotton, while, TYLCV transcripts increase in whiteflies transferred to cotton and allowed to feed on cotton for up to seven days. Plant viral transcriptional activity within the insect vector was associated with lower viral DNA titer in the plant and different viral DNA allocation patterns within the whitefly. This is the first quantitative report of a plant DNA virus transcriptional activity in an insect vector, and correlation of this activity with viral DNA abundance in the plant.

Identification of the Genes Associated with Tomato Irregular Ripening using Microarray Technology

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Tomato irregular ripening (TIR) disorder has been associated with silverleaf whitefly (SLW) (*Bemisia argentifolii* Bellows and Perring) feeding. Symptoms of this physiological disorder do not appear on tomato foliage where whitefly feed, but appear as an uneven fruit color development. Plants infested with SLW can develop fruit with external longitudinal white or yellow streaks resulting in unmarketable fruit. TIR internal symptoms consist of a lack of internal coloring of the fruit, thereby reducing post harvest quality. The mechanism(s) associated with the development of TIR has not been identified. Gene array technology was applied to TIR as a model system. Our objective was to study gene expression over time in tomato plants subjected to a moderate infestation of whitefly and resulting in the development of TIR. Multiple plant harvests of old and young leaves, stems, roots, flowers and fruit from tomato with and without whitefly were processed for nutritional analysis and RNA extraction. RNA was labeled and hybridized to the gene array membrane to determine which genes SLW feeding influences. Plants infested with whitefly were larger, produced more leaves, flowers and fruit, but significantly less fruit ripened compared to plants without whitefly. Plant nutrition and RNA profiling results will be presented.

The *Bemisia tabaci* Functional Genomics Project

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We propose the first genomics project on a hemipteran insect pest to agriculture, the whitefly *Bemisia tabaci*. This insect transmits many viruses to a variety of economically important crops in the USA, Israel, and worldwide. The B type (synonym *B. argentifolii*) is resident to Israel, and has invaded the Southern USA, reaching epidemic proportions. Despite its extreme economic importance, there is little molecular genetics information on *B. tabaci*. This project proposes the development and public presentation of a *B. tabaci* expressed sequence tag (EST) library and associated cDNA high-density arrays, and to use the arrays to analyze how begomoviruses influence the whitefly transcript profile. The cDNA libraries will be constructed by pooling populations of B biotype selected before and after they acquire mono partite (*Tomato yellow leaf curl virus*) and bipartite begomoviruses (*Tomato mottle virus*). Genes whose transcript abundance changes in response to acquisition and circulative movement of begomoviruses through the whitefly will be identified using array-based hybridizations. This project combines expertise in *B. tabaci* biology, plant-virus-vector relationship, genomics and bioinformatics, and is presented with the knowledge that understanding the molecular mechanisms underlying the biology of *B. tabaci* is a prerequisite for the creation of new strategies to curb the disastrous effects of this insect on many crops worldwide. Preliminary EST results will be presented from the various whitefly libraries.

Plant-mediated effects of drought, nutrient and heavy metal stress on a grass miner

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We investigated the predictions of the plant stress and the plant vigour hypothesis with the grass miner *Chromatomyia milii* (Diptera: Agromyzidae) on drought, nutrient and heavy metal stressed *Holcus lanatus* plants. Plant stress intensity was measured by recording plant growth. Plant growth was positively related to insect preference in the water and the heavy metal stress experiments. This corresponded with a decrease in performance of *C. milii* with increasing plant stress intensity. Our nutrient treatments had little effect on plant growth, except for a decrease in plant biomass when an excess of nutrients was supplied. Offspring survival was also lower on the *H. lanatus* plants that were stressed by an excess of nutrients. The decrease in performance on the water stressed plants was mainly caused by self-pruning of the *H. lanatus* plants. Heavy metal stressed plants were unsuitable resources because of an increasing accumulation of foliar cadmium, and a decreasing foliar carbohydrate concentration, with increasing heavy metal exposure. In conclusion, *C. milii* always showed a negative response to stressed plants. Yet, the mechanistic explanation for the decrease in performance on stressed plants differed among stress types.

A standardized method for monitoring solitary bee communities

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Bees are the principal pollinators of flowering plants in most terrestrial ecosystems of the world and arguably no taxonomic group has been more important for the evolution and function of these ecosystems. Bee activity translates broadly and directly to human well-being. In North America alone, estimates of the value of bees (natives and the introduced honey bee) as pollinators of crops ranged between $4.6 and $18.8 billion. Despite their well recognized economic and ecological value, we lack basic information on the global, regional, and local patterns of diversity and population trends of this species rich and ecologically important group. To begin to fill this gap, we have developed and tested a simple sampling protocol (http:\\online.sfsu.edu\~beeplot) that requires low cost equipment available worldwide. Our protocol is usable by scientists of many interests, as well as amateurs and school groups. Data generated using this protocol will be useful for 1) conservation planning and strategy development 2) local, regional and national assessments of species richness; and 3) evaluations of population trends in different habitat types (e.g. agricultural versus wilderness sites). This collaborative effort will allow us to detect differences in species richness and population trends among biomes, ecoregions, habitats, communities, and plots.

The diversity and distribution of Coleoptera in three different plant communities on Mt. Tangkubanparahu, West Java, Indonesia

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Situated in Java Island, Indonesia, Mt.Tangkubanparahu possesses many interesting features, among others are floristic composition, elevation, and level of human disturbance, which undoubtedly affect the abundance and diversity of the fauna including those belong to the order Coleoptera. The study on diversity and spatial and temporal predominance of Coleoptera was conducted in three different plant communities namely Schima-Astronia forest(1600 m asl),pine plantation(1300 m asl) and agroforestry area (1200 m asl) using pitfall traps. The total abundance of Coleoptera in the three communities was 1284 individuals comprising 127 species from 22 families. The highest abundance was found in agricultural site (510 individuals) followed by in mixed forest (506 individuals) and in pine forest (268 individuals). The highest species richness was found in mixed forest (77 species) followed by in pine forest (55 species) and agricultural site (58 species). The index of diversity (H') was the highest in mixed forest (3.67) while that of pine forest and agricultural site were 3.30and 3.04 respectively. Coleoptera were found most abundant in mixed forest and in pine forest in November (161 and 94 individuals respectively) while in agricultural site in December (224 individuals). The highest temporal index of diversity in mixed forest and pine forest were found in November (H'= 3.21 and 3.10 respectively) while in agricultural site in December (H' = 2.99).

Colonisation of terrestrial bromeliads by insects in the Atlantic rain forest of Santa Catarina, Brazil

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Bromeliads offer suitable environments for the reproduction of large numbers of animal species because their central leaf rosette forms small water bodies where aquatic insects live, while the lateral leaf axils hold decomposing organic material inhabited by terrestrial organisms. In order to better understand the dynamics of the animals that make use of bromeliads we performed a study on the colonization of *Aechmea lindenii* by terrestrial insects. Plants were manipulated by filling the central tank and the leaf axils with sterilized humus. For 10 months, a sample of 5 plants was collected every 4 weeks and all insects associated with them were recorded. 5 unmanipulated plants of the same species served as a control. During the same period, the soil fauna of patches close to the bromeliads was sampled by Winckler extraction. Both bromeliad groups contained about equal amounts of ants (75%). For the other arthropod groups, spiders and beetles dominated in manipulated bromeliads, while the controls contained about 50% aquatic beetles and flies. Our data suggest that the plants that were transformed into a more terrestrial habitat were successively colonized by a typical terrestrial arthropod fauna that presents similarities to the soil fauna as analyzed by the Winckler extraction.

The role of an adventitious mutualism and water stress in population outbreaks of scale insects

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Outbreaks of pest insects are often preceded by stressful weather conditions, where stressed plants become susceptible to herbivores due to altered nutritional quality or defensive chemical concentrations. Some insect species also increase in abundance via beneficial interactions. We studied the explosions in density by the invasive yellow crazy ant, *Anoplolepis gracilipes*, on Christmas Island, which were concurrent with a rapid increase in populations of introduced/cryptogenic honeydew-producing scale insects. We tested the combined effect of plant water stress and attendance by *A. gracilipes* on the success of populations of the lac scale, *Tachardina aurantiaca*, on seedlings of rainforest species. Plant water stress had no significant effect on the abundance *T. aurantiaca* crawlers. Nor did stressed plants affect the abundance or establishment of adult females. However, the presence of *A. gracilipes* had a highly significant, positive effect on all life stages. After 2 months, densities of adult females increased 4-fold on seedlings with ants compared to those without ants. Our results suggest higher rates of crawler establishment and increased survival throughout development in association with an introduced ant mutualist, independent of plant water stress. Establishment of adventitious mutualisms following alien species introductions may play a pivotal role in accelerating population explosions and concomitant impacts of invasive insects.

Nightshade plants attract aphid vectors and harbor potato viruses

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Hairy nightshade, *Solanum sarrachoides* is a solanaceous weed abundantly distributed in potato ecosystems of Idaho, the largest potato producing state in the U.S. It is a preferred host for green peach aphid (GPA) *Myzus persicae*, considered the most effective vector of potato leafroll virus (PLRV). Our previous laboratory experiments confirmed that GPA can transmit PLRV to hairy nightshade and that non-viruliferous aphids can become viruliferous after feeding on infected nightshades. Six percent of field-collected nightshade plants in 2002 were found to carry PLRV and in few cases other viruses such as PVY and PVX. We investigated whether PLRV-infected nightshades would attract GPA and also how this attraction would compare with the one produced by infected potatoes. Field trials were conducted to study the role of *S. sarrachoides* on vector distribution and PLRV epidemiology. Aphid counts and disease progression were monitored at weekly intervals. Results revealed that plots with nightshades recorded an increased number of aphids and also had higher number of PLRV infected potato plants. A preference study revealed increased preference of winged GPA to PLRV-infected *S. sarrachoides*. Our findings thereby suggest that *S. sarrachoides* might play a major role in the epidemiology of PLRV in the potato ecosystem.

Annual cycle of the African rice gall midge in southwestern Burkina Faso: host-plant relationships

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The African rice gall midge, *Orseolia oryzivora*, is a major pest in Burkina Faso. The present study was carried out during 4 consecutive crop seasons from 1999 to 2001 in three rice irrigated schemes in the southern part of the country. It aimed at monitoring the pest damage and its immature stage populations through observations on growing rice in farmer fields, on rice ratoons after harvest and on wild rice around rice fields. The pest survival in absence of rice was due to rice ratoons and wild rice, *Oryza longistaminata*. However, rice crop was the preferred host plant as over 15 % of galls were observed during wet seasons while damages and larval populations were very low in dry seasons. These results are important steps for the development of IPM for the pest control with regard to its host plant relationships.

Acridids as a model system for investigating plant nutrient-secondary metabolite interactions and herbivore foraging behaviour

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Insect herbivores forage to acquire nutrients, but they rarely encounter plants that contain nutrients in optimal ratios. Additionally, many plants contain secondary metabolites (PSMs) that are toxic or reduce food utilisation. Thus the challenge insect herbivores regularly face is one of meeting their nutritional requirements while simultaneously limiting the intake of detrimental compounds. Locusts have proven to be a useful model organism for investigating nutrient regulation under a number of different scenarios, and they are now being used to disentangle the influence of PSMs and nutrients on insect herbivore foraging behaviour. In this talk I present results from a series of experiments that examine feeding responses by locusts to foods containing different nutrient compositions and various levels of carbon- (tannic acid) or nitrogen-based (gramine) PSMs. The results demonstrate that the mode of action for a particular PSM can vary depending on a foods nutritional profile, and that an herbivore's response to a PSM can be modified depending on the chemical composition of the foods that are available. I conclude the talk by discussing the broader implications of these findings, specifically how they relate to plant defence theory and models of foraging behaviour.

Insect herbivore learning: what can grasshoppers teach us?

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Numerous examples exist of insect herbivores developing learned aversions towards particular foods. In contrast, little is known about the extent to which insect herbivores learn about their foods following positive experiences. Grasshoppers are ideal subjects for such questions because of their biology and amenability to laboratory conditions. In this talk I present a novel protocol, based on a Y-maze, which is specifically designed for studying learning and choice behaviour in grasshoppers (*Schistocerca gregaria*, the desert locust is used in this study). Results from the first three experiments using this system demonstrate that *S. gregaria* can readily learn to associate colours and odours paired with the larger of two food rewards, and that it reaches the larger reward option faster. In the last experiment, the same protocol was used to examine choice rules related to foraging decisions. Results here suggest that feeding decisions were not exclusively based on the intrinsic properties of the options. Rather, they show that locusts preferred options that had a history of yielding higher fitness gain. I conclude the talk by discussing the benefit insect herbivores gain through forming positive learned associations, and the enormous potential of using grasshoppers as a model system for investigating learning in general.

Temporal variation in nectar production in two sympatric species of *Schefflera* (Araliaceae) – A strategy to overcome competition for pollinators?

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During our studies on the flowering phenology of shade plants in coffee plantations, we came across two sympatric species of *Schefflera* (Araliaceae), viz., *S. venulosa* and *S. wallichiana* in the Western Ghats of India. These two species flower synchronously in the month of May and attract a large number of insects including honey bees. Of the bee species visiting *Schefflera* flowers, the Indian hive bee, *Apis cerana* was predominant (53%) followed by the stingless bee *Trigona* irridipennis (15%). Inflorescences of *S. wallichiana* were significantly larger with more number of flowers ($x = 4160.5 \pm 2169$; $n = 40$) compared to that in *S. venulosa* ($x = 2075 \pm 216$; $n = 40$). *Schefflera wallichiana* flowers secreted significantly higher quantities of nectar compared to those of *S. venulosa.* The two species of *Schefflera* exhibited a clear temporal segregation in nectar production, there by attracting pollinators in different times of the day. In *S. venulosa* nectar production was in the morning hours (between 0830 and 1130 hrs), while that in *S. wallichiana* it was during the afternoon hours (between 1300 and 1600 hrs), which resulted in shifting of the pollinators from *venulosa* flowers in the morning to *wallichiana* flowers in the afternoons.

Do herbivore arthropods benefit from vectoring plant-viruses?

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Although much study has been devoted to mechanisms by which plant pathogens become vector-borne, it is still unclear why herbivorous arthropods should vector the pathogen. To address this question, we use a system consisting of *Tomato spotted wilt virus*, its thrips vector (*Frankliniella occidentalis*) and pepper plants. We apply a combination of plant and vector treatments to discriminate between indirect (*i.e.* through the shared host plant) and direct effects of virus infection on the thrips vector. The main positive effect of the virus on the vector is indirect. The attack of plants by the vector results in a reduction of plant quality for the vector, but virus infection of plants alleviates these effects of vector-induced plant defenses. Hence, it is advantageous for the vector to infect plants with virus or to develop on infected plants. Mechanical inoculations allow for testing the effect of the virus without herbivory, indicating a positive effect of virus infection of the plant further than overcoming induced plant defenses to herbivory. The benefit of virus infection for potential vector's performance is regardless of their ability to transmit the virus. The consequences for the food web of the presence of the virus in the system are discussed.

How species influence ecosystem function: grasshoppers influencing rangelands

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Conservation of biodiversity is often justified with claims that particular species are critical for maintaining ecosystem processes, such as nutrient cycling and net primary production (NPP), i.e., ecosystem structure controls function. Experiments in western Montana (USA) rangelands indicate that grasshoppers can accelerate nutrient cycling and thereby increase NPP by as much as 20% in some areas, or decelerate nutrient cycling and thereby decrease NPP by as much as 10% in other areas. In some areas one grasshopper species can increase NPP, while another grasshopper species decreases NPP. However, in other nearby areas at the same elevation and with the same plant species, the same two grasshoppers can have reverse effects on NPP. These observations ultimately rely on grasshopper feeding behavior. When grasshoppers increase NPP, they preferentially feed on the plants producing slower decomposing litter. However, when grasshoppers decrease NPP, they preferentially feed on plants producing faster decomposing litter. Each grasshopper's feeding preferences can change as plant characteristics vary in relation to the grasshopper's physiology in each area. Finally, predation on herbivores is often thought to enhance NPP. Our results indicate that this is only true if herbivory diminishes NPP. However, predation will decrease NPP if herbivores increase NPP.

Multitrophic gene-family interactions: cytochrome P450 monoxygenases in the wild parsnip/parsnip webworm interaction

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Genome sequencing has yielded an embarrassment of riches with respect to certain gene families; insect genomes, e.g., can contain in excess of 100 cytochrome P450 genes. Because these genes play a role in mediating interactions among organisms, understanding the evolution of P450s within a genome necessitates characterizing trophic interactions at the molecular level. The interaction between wild parsnip (*Pastinaca sativa*) and the parsnip webworm (*Depressaria pastinacella*) is mediated by furanocoumarins: both the detoxification of furanocoumarins by webworms and their biosynthesis by parsnips involve cytochrome P450-catalyzed reactions. Thus, observed phenotype matching between detoxification profiles in the insects and furanocoumarin production in the plants reflects underlying variation in, among others, P450 genes in both insect and plant. In an initial effort to characterize these gene-family interactions, four cDNAs were cloned from webworm midguts. One of these P450s, CYP6AB3, is inducible by furanocoumarins and baculovirus-mediated expression studies confirm its role in metabolizing these compounds. Structural superpositioning of molecular models of this P450 and two CYP6B furanocoumarin-metabolizing *Papilio* P450s reveals high spatial similarity of elements within the catalytic sites of these enzymes, suggesting convergence across the guild of insects specializing on furanocoumarin-containing plants. Characterization of promoter sequences in P450 genes reveals both unique elements that may promote specialization and more generalized elements that may allow insects to respond preemptively to plant defense. The existence of a significant third trophic level, in the form of a specialist encyrtid parasitoid, in some parts of the range of parsnips and webworms indicates that selection by and on yet another suite of P450 genes can influence coevolutionary responses in this interaction.

Investing in growth vs. defense by plants under attack in a multitrophic context.

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Plants are faced with a limited amount of resources to invest in different functions such as growth, reproduction and defense. This often results in trade-offs between investment in defense and other fitness-enhancing traits. Optimal investment in secondary metabolites involved in direct chemical defense therefore depends on the costs and benefits associated with different levels of these defense compounds. Costs of both constitutive and induced defense have been documented in numerous systems. But clearly, in order to understand optimal levels of direct chemical defense compounds in natural plant populations, the magnitude of costs and benefits should be considered in a multitrophic context, because direct chemical defense may show compatible or conflicting associations with indirect defense (attraction and efficacy of natural enemies of herbivores). First, direct chemical defense can have cascading effects, affecting availability and quality of insect herbivores to their parasitoids and predators. Second, positive or negative genetic associations between direct and indirect defense may arise from shared biochemical pathways of allelochemicals and volatiles involved in parasitoid attraction, requiring insight into the underlying mechanisms. I review some of the interactions between direct and indirect defense and its potential consequences for selection on defense in natural plant populations.

Effect of transgenic resistance to *Barley yellow dwarf* luteovirus in wheat on virus-vector relationships

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The life history of the bird cherry-oat aphid, *Rhopalosiphum padi* (L.), was examined on plants of a transgenic genotype and an untransformed wheat variety infected with *Barley yellow dwarf luteovirus* (BYDV), or non infected. Transgenic plants expressed the coat protein gene from BYDV-PAV and showed moderate resistance to the virus. Based on nymphal development period, length of reproductive period, total fecundity, and intrinsic rate of increase, virus-infected transgenic plants were inferior hosts for *R. padi* compared to virus-infected untransformed plants. However, non-infected transgenic plants were superior hosts for *R. padi* than non-infected untransformed plants. The effects of different acquisition access periods (AAP) on the transmission efficiency of BYDV by *R. padi* following feeding on transgenic or untransformed wheat, were also examined. Transmission efficiency increased with an increase in AAP in both, transgenic and untransformed wheat. However, aphids were less efficient vectors following feeding on virus-infected plants of the transgenic genotype compared to the untransformed variety. Our results show that transgenic virus resistance in wheat can indirectly influence *R. padi* life history and virus transmission efficiency.

Flower visitor assemblages associated with rainforest trees

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The assemblages of insects which visit particular flower species within an ecosystem may reflect a predictable, organised, perhaps co-evolved, set that is responding to the particular inflorescence based on the colour, morphology, phenology or chemistry of the plant concerned. Alternatively the assemblage may represent a set that is assembled more or less by chance from the overall canopy fauna. Testing the specificity of flower visitors may be carried out in one of two ways; first, by monitoring the changes in visitors to a species across a number of seasons; and, second, by comparing visitor assemblages to co-flowering species within a population. Using several independent methods we have studied the visitor assemblages associated with ten different canopy plant species within the lowland rainforest of North Queensland. Even an analysis at the ordinal level shows that there are substantial, statistically significant differences across plant species. It seems likely that visitor assemblages represent a predictable response to particular plant species in most cases. We did however find evidence that the visitors to a number of plant species' flowers reflected general canopy insect 'traffic'. Uncertainty remains about the levels of host specificity involved, and the underlying variability in assemblage structure both within and between years.

Responses of grasshoppers to livestock grazing: habitat manipulation by mammalian herbivores

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Little experimental work has examined the ecological interactions between livestock and grasshopper herbivores, and the effects of differing patterns of herbivory by livestock on grasshopper population dynamics have not been fully examined in western US rangelands. Grasshopper habitat manipulation in the form of livestock herbivory has been shown to impact grasshopper population dynamics. Mammalian herbivory can modify factors important for grasshoppers such as plant species composition, host plant quantity or quality, plant cover and microhabitat conditions. A multi-year cage experiment was conducted to examine how the presence, timing and intensity of sheep herbivory affected grasshopper population dynamics and vegetation. There were few initial treatment differences in grasshopper densities between the timing and intensity of sheep grazing, although grazing affected the biomass and quality of vegetation available for grasshoppers. Grasshopper population densities and community composition were assessed in pasture scale livestock grazing experiments. The results from the pasture scale studies indicate that differences in livestock grazing can affect grasshopper population dynamics, but treatment differences may not be apparent when grasshopper population densities are very low. In addition, habitat differences resulting from livestock herbivory may not occur within a single season.

Insect views on plant structural complexity

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What are the relative impacts of plant architecture and resource distribution on the movement and searching efficiency of males searching for females, of herbivores searching for oviposition sites, and of predator searching for prey in plant canopies? How far is movement function of the architecture and stimuli stemming from the resource? How shall we deal with canopy-to-canopy architectural variations? All these questions require quantification of (i) the architecture of the environment, (ii) the distribution of resources within it and (iii) the intrinsic rules of movement. The appropriate analytical framework is based on random walks in random environments. Using a large data set of the apple fruitfly *Rhagoletis pomonella* foraging in apple trees, we built a foraging model that considers the effect of canopy structure and the presence of fruits for oviposition. We then estimated the sphere of attraction of a single fruit by comparing predicted and paths. Finally, we present the latest computational advances required in modelling realistic canopy architecture using growth units and multi-scale structures. These techniques offer the prospect of dealing efficiently with the difficult problems of comparing and simulating canopy architectures, both in topological and in three dimensional geometrical terms.

Foliar nutrients and induced susceptibility: genetic mechanisms of Douglas-fir resistance to western spruce budworm defoliation.

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We conducted greenhouse defoliation experiments with clones of Douglas-fir over three years to assess the role of foliar nutrients as a resistance mechanism to western spruce budworm defoliation. The grafted clones were derived from mature trees (i.e., ortets) that showed resistance or susceptibility to budworm defoliation in the forest. Current-year foliage was analyzed for concentrations of nitrogen, sugars, and eight minerals. We computed rank correlations between foliar nutrient levels in the ortets and their corresponding clones to test the null hypothesis that foliar chemistry does not have a genetic basis. Foliar concentrations of sugars and phosphorus were under genetic control to some degree, but concentrations of other nutrients were not. We used analysis of variance to test the null hypotheses that foliar chemistry does not change in response to budworm defoliation. We rejected the hypothesis for sugars, P, potassium, manganese, and zinc; defoliation by the budworm changed levels of these nutrients, and had divergent effects on concentrations of P, K and Zn in resistant compared to susceptible clones. We concluded that induced susceptibility, whereby defoliation alters foliar nutrients to make trees more favorable for insect feeding, appears to be an important determinant of Douglas-fir resistance to the western spruce budworm.

Influence of diet chemistry, anatomy, and biomechanical properties on Australian plague locust performance

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The majority of locust nutritional physiology studies have been undertaken using artificial diets. Comparison of dietary utilisation and growth and development of the Australian plague locust on two contrasting grasses showed that existing models could not be used to predict locust performance. Following rain, Australian plague locusts develop in Mitchell grasslands which are dominated by perennial Mitchell grass and annual Button grass. Both grasses were similar in terms of the major chemical constituents. However, they differed anatomically, with Button grass having *c.* twice the specific leaf area and water per unit dry matter. Plant anatomy not only affected diet digestibility and post-digestive processing costs, but also influenced the rate of nutrient assimilation. When the barrier effect of the indigestible cell wall was removed by grinding, diet digestibility doubled, resulting in increased dry matter assimilation. This, together with reduced metabolic costs of converting these nutrients to biomass, increased growth. Early instars were able to digest and grow on either diet equally, but the later instars consuming Mitchell grass experienced reduced growth and increased instar durations. This appeared to be due to reduced consumption resulting from slower nutrient assimilation rather than lowered diet digestibility or increased metabolic costs associated with food processing.

Visual/Mechanosensory interactions and the evolution of insect behavior

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A major goal of neurobiology is to understand the design principles of cells and circuits that control behavior. Orthopteroid Insects provide ideal models for understanding the principles underlying the design of uniquely identifiable brain cells related to orienting behavior. Specific brain cells ("DMIs") of cockroaches trigger evasive turns in response to touch [abrupt antennal contact] from predators. The antennae of cockroaches are under visual guidance, apparently in part by other specific brain cells ("DCMDs") that were originally described in locusts. Our preliminary evidence suggests that cockroaches, crickets, locusts and mantids all possess a DMI / DCMD ensemble, and that these two interneuron systems operate in a coordinated way to control directional motor behaviors specific to each species. Comparative analyses on the four orthopterod species mentioned suggest the hypothesis that across these species, the sensory encoding properties of these interneurons have been conserved, but that their role in behavior has been adaptively altered by localized cellular changes in motor outflow organization at cerebral and/or thoracic levels of the CNS.

Developing novel resistance to *Helicoverpa* spp. in chickpeas

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Helicoverpa species are highly polyphagous, and are highly damaging pests of most agriculturally important crop plants. The development of cultivars with resistance to these pests is an economically and ecologically promising solution, but the low levels of genetic resistance in many crop species has led to the examination of wild relatives as possible sources of resistance to this insect. Screening of several wild *Cicer* accessions has led to the identification of putative resistant germplasm that could be crossed into cultivated chickpea varieties. In addition, an understanding of the genetic basis of host plant selection by these polyphagous pests may lead to novel approaches to control them in the future. We investigated the quantitative genetics of adult preference and larval performance on different chickpea genotypes, at different life stages, as well as the genetic correlations between traits. We found high levels of additive genetic variation in the population for its ability to feed on both susceptible and moderately resistant chickpea genotypes. Furthermore, we found significantly higher levels of variation in neonates than in the older larvae. The implications of these findings are discussed in relation to the potential of *Helicoverpa* to overcome host plant resistance in agriculturally important crops.

Life-history differences in the genus *Lipara* (Diptera) on their common hostplant, the grass *Phragmites australis*

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The flies of the genus *Lipara* (Diptera; Chloropidae) are strict monophagous gall formers on the common reed, *Phragmites australis* (Poaceae). Some species induce clearly defined gall chambers while other do not. The *Lipara* species are not homogenously distributed in their common habitat. Each species' larval survival and populations are uniquely correlated with reed shoot diameter (a measure of plant vigor). The ovipositing females are able to discriminate between shoots of different quality. The different species also support different parasitoid guilds. At least one species acts as a kleptoparasite in the galls of other *Lipara* species. Kleptoparasitism in this system is mediated by the host plant and by the principal parasitoid. During the evolution of the genus *Lipara* a trade-off between obtaining the necessary food supply and growth dependent host plant resistance has been a major force in structuring the life cycle strategies.

Spatial heterogeneity in monoterpene concentrations of native pine as a driver of insect herbivore distribution and abundance

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We investigated whether the genetic diversity of structural species in native ecosystems can control the diversity of other taxa. This study focused on insect herbivores associated with Scot's pine (*Pinus sylvestris*) and tested hypotheses that variations in the monoterpene chemistry of individual pine trees will affect herbivore diversity and performance, and the quality of litter fall available to soil organisms.
Three methods were used;
Funnel collectors to sample litter fall from the crowns of 45 pine trees, selected to represent monoterpene variability across the forest.
A bioassay measured growth rates and survival of pine looper moth caterpillars, using net enclosures in the crowns of 30 pine trees.
Measurements of the numbers of ants climbing pine trees and the plant and arthropod material they transported to the ground.
Numbers of phytophages were related to the concentration of certain monoterpenes and tree structure and location. The growth of pine looper caterpillars was related more to chemistry than physical parameters. Ants transported significant material from pine crown to ground and removed phytophages other than aphids from the crowns. We discuss the contribution of genetic diversity for phytophage diversity and the broader ecological consequences.

Induced plant defences – using *Arabidopsis* as model for linking ecology with molecular biology.

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In their defence against pathogens, herbivorous insects and mites, plants employ many induced responses. One of these responses is the induced emission of volatiles upon herbivory. These volatiles can guide predators or parasitoids to their herbivorous prey, and thus benefit both plant and carnivore. This use of carnivores by plants is termed indirect defence and has been reported for many plant species, including elm, pine, maize, lima bean, cotton, cucumber, tobacco, tomato, cabbage and *Arabidopsis thaliana*. Herbivory activates an intricate signalling web and finally results in defence responses such as increased production of volatiles. Several components of this signalling web are known, such as the plant hormones jasmonic acid, salicylic acid and ethylene. Information on the molecular genetics as well as the phenotypic changes that are induced by herbivory can be exploited to design experiments that aim at assessing the effects of plant traits on the interactions with members of the food web in a multitrophic context.

Latex: mode of action, efficacy, and insect response

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Over 5% of all plant species possess latex canals. The plants sequester adhesives and poisons within these pressurized delivery systems that release liquid latex immediately upon injury. Herbivores biting a leaf have their mouthparts coated instantly with noxious exudates that often become increasingly sticky before eventually hardening. Limitations of latex defenses include a vulnerability to herbivores capable of rupturing canals before feeding distal to the cuts in an area of reduced exudation. Insects sever canals with vein-cutting, trenching, and girdling behaviors that have evolved repeatedly in diverse insect groups. I will discuss how latex defenses affect insect foraging behaviors, herbivore distribution on individual plants, and host range. These studies document that latex exudation can harm both mandibulate and haustellate herbivores, as well as the natural enemies that prey upon them.

Bioprospecting with a caterpillar probe: identification of trenching stimulants

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Over 100 insect species sever veins or cut trenches in leaves before feeding beyond the cuts. The insects thereby depressurize defensive canal systems and prevent exudation of latex, resin, or other fluids during feeding. Whether the cuts function to reduce insect exposure to noxious chemicals in exudates, to adhesives, or both has been debated. Using a trenching bioassay with cabbage loopers, *Trichoplusia ni*, I have identified three compounds that elicit trenching: lactucin from lettuce latex, myristicin from parsley, and lobeline from cardinal flower. All three compounds affect the nervous system of mammals. Other neuroactive chemicals, including some neurotransmitters, insecticides and drugs also trigger trenching. The known toxicity of many of these compounds supports the hypothesis that cabbage loopers trench specifically to reduce their ingestion of noxious exudate constituents. Trenching assays thus have potential for directing the isolation of novel plant compounds with insecticidal properties and potential applications in pest management or pharmacy.

Aphid dispersal patterns from point sources: implications for the introduction and spread of viruses in crops

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The epidemiology of plant virus outbreaks is driven by the behaviour of the vector and the magnitude and disposition of the virus source. Controlling the damage caused by insect-transmitted viruses in annual cropping systems is greatly assisted by an understanding of the movement patterns of the vectors. In this paper, we describe field experiments designed to monitor the short-distance dispersal patterns from point sources of two species of cereal aphids. These experiments showed that most aphids disperse only a short distance (<5-10m) from their source. The relative probability of dispersal to host plants outside this range does not decrease greatly for distances up to 200m, if one factors out the dilution effects caused by increases in dispersal area. Our first year of experiments using anti-feeding pesticide barriers on the edge of fields indicated that a 25m-width barrier was insufficient to affect aphid immigration rates. Independent experiments examined the effectiveness of non-host barriers at diminishing virus introductions into crops by thrips and aphid vectors. Future experiments will combine these approaches, monitoring virus-infected aphid dispersal and population increase while also monitoring virus spread into the affected crop.

Response of aphid vectors of plant viruses to volatiles from virus-infected plants

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During host selection, insect herbivores potentially respond to intraspecific variation in host plant quality, including that resulting from infection by disease–causing organisms. For example, virus-infected plants sometimes are superior and preferred hosts for the aphids that vector the infecting viruses. We have shown that virus-induced changes in headspace volatiles (VIVs) of infected plants influence host selection behavior by two aphid species in three pathosystems. The green peach aphid, *Myzus persicae*, is more strongly arrested by VIVs from *Potato leafroll virus*-infected cultivated potato, *Solanum tuberosum*, or wild *S. sarrachoides,* than by the headspace volatiles from virus-free plants of these species. The bird-cherry oat aphid, *Rhopalosiphon padi*, responds similarly to VIVs from wheat plants infected with *Barley yellow dwarf virus.* Response to VIVs not only is beneficial to the aphids, which are aided in locating superior hosts, but also potentially increases the rate of spread of the viruses. In such an apparently mutualistic relationship, the signals and responses to them potentially are shaped by natural selection to optimize accrued benefits. With this general hypothesis in mind, we examine the variation in VIV production and aphid responses to this variation throughout the disease cycle in the Potato-PLRV-*M. persicae* pathosystem.

Plant morphology at different spatial scales and insect-plant interactions

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The physical form of plants influences insect-plant interactions at several spatial scales. At relatively small scales, plant morphological features such as trichomes and surface waxes influence the ability of phytophagous and carnivorous insects to cling to and move over plant surfaces. The requirement to cling successfully to plants has been met through modifications of the attachment devices and behavior. These include variation in the anatomy of pretarsal claws and legs, the form and extent of adhesive pads and setae, the ability and propensity to construct silken platforms for resting, and behavioral accommodations for walking on plant surfaces. Adaptations evidently have also occurred in the ability of insects to obtain adhesive or frictional attachment to leaf surfaces, although these are as yet little understood. At larger scales, the plant soma presents a complex three-dimensional environment that insects must negotiate to find resources. Foraging in the plant canopy requires effective movement rules for finding resources efficiently. Potential effects of plant architecture on insect-plant interactions include multitrophic and coevolutionary interactions. In the talks that follow in this symposium, we will hear from scientists examining the effect of plant physical features at different spatial scales on insect-plant interactions. As we hear these talks, let us also consider how plant morphology at different spatial scales might interact to influence insects and shape the adaptive syndrome of insects foraging on plants. The influence of plant structure on insect ecology perhaps can be better understood through such a multi-scaled perspective.

Insect feeding behaviour and the transmission of plant viruses

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In the last years significant progress has been made on the specific feeding behaviour events associated to the acquisition and inoculation of plant viruses by their insect vectors. Most efforts have concentrated on aphid-transmitted viruses and particular behavioural processes leading to the transmission of non-circulative and circulative viruses have been identified. This paper will review on how viruses and other plant pathogens have adapted to exploit particular feeding habits of their insect vectors to spread from plant to plant. In particular, some phloem-restricted viruses (e.g. Luteoviridae and Begomovirus) use phloem feeders such as aphids or whiteflies for dispersal after circulation through the insect's hemocoel and excretion through the saliva. Others (e.g. Closteroviridae) are also acquired from the phloem but remain attached to the mouthparts of the vector until subsequent feeding on the phloem tissue of a healthy plant. Xylem-restricted bacteria are vectored by some Cicadellidae that penetrate the plant tissue intracellularly to feed in the xylem vessels. Other viruses (e.g. Caulimoviruses) are transmitted after several intracellular stylet punctures before starting phloem salivation or feeding. On the other side, there are viruses (e.g. Potyvirus) that have a very ephemeral interaction with their vector and are transmitted to the epidermis after single intracellular stylet punctures and are lost well before the vector reaches the vascular tissues.

Transmission of *Cauliflower mosaic virus*: **behavioural events associated to the inoculation process**

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"The transmission efficiency of *Cauliflower mosaic virus* is highest when the virus is acquired from the phloem, but information on the relationship between the different behavioural events associated to the inoculation phase is lacking. For a better understanding of the transmission mechanism of CaMV, we used the electrical penetration graph (EPG) technique to assess aphid feeding behaviour during the inoculation of CaMV by *Brevicoryne brassicae* L. and *Myzus persicae* Sulzer. to *Brassica rapa* (cv. Just Right) plants. After 8 hours of acquisition on CaMV infected plants, aphids were placed on test plants and removed immediately after specific behavioural events were recorded, and then transferred to a second test plant. Aphids transmitted CaMV with higher efficiency when five or more intracellular punctures were recorded or when they probed for five minutes on the test plants than when a single intracellular puncture was produced. No increase in the transmission rate of CaMV was observed when aphids reached the phloem phase. Interspecific differences were found in the retention and persistence of CaMV in its vectors. *B. brassicae* could retain the virus after several intracellular punctures were recorded whereas *M.persicae* lost the virus after performing the same number of punctures.

Intra- and inter-population correlations between growth rate, survival, and nutritional indices of *Melanoplus sanguinipes* (Orthoptera: Acrididae) on low quality diets.

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Growth, survival, food consumption and assimilation were examined in grasshoppers from two different populations. One from Idaho (ID) and the other from Alaska (AK) where summers are cool and moist. When reared on a lettuce/bran diet, the ID grasshoppers develop more slowly and with greater variance in growth rates than the AK grasshoppers. For the experiment, grasshoppers were reared through the 4th and 5th instars on one of two artificial diets containing 0.7 or 2.6%N. The ID grasshoppers were split into two groups, those that attained 4th-instar in 15 to 17 days (ID-Fast) and those that required more than 19 days to reach 4th-instar (ID-Slow). The AK grasshoppers reached 4th-instar in 13 to 15 days. No individuals of the AK or ID-Fast groups survived to adult stage on the 0.7%N diet, whereas 58% of ID-Slow survived. On the 2.6%N diet, 47% of the AK survived to adult stage, and all individuals of both ID groups survived. Relative consumption rates of ID-Slow were greater than the AK grasshoppers. Assimilation rates in the ID-Slow were greater than for ID-Fast and AK grasshoppers. It appears that individuals that grow more slowly under optimal conditions, are better able to tolerate a poor quality diet through greater consumption and assimilation of food.

Silk production in the Hemiptera

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Research on leafhoppers, *Kahaono montana* Evans (Cicadellidae: Typhlocybinae: Dikraneurini), living under silken shelters on the vertical leaves of species of *Eucalyptus* (Myrtaceae) has shown that these shelters are produced by the leafhoppers themselves. The silk has some chemical similarities to spider silk but very different physical properties. Feeding damage by the leafhoppers is limited to the area covered by the shelters. Liquid excrement expelled by the leafhoppers accumulates on the fibres and blackens through the growth of sooty moulds. Evidence will be presented suggesting that these shelters function to trap and retain heat to protect leafhoppers living in cooler regions of Australia. There are indications that at least one other species of Dikraneurini also produces silk.

Creating habitat for native bee pollinators in highly modified landscapes

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The publication of "Forgotten Pollinators" in 1996 by Buchmann and Nabhan has raised much awareness about the decline of pollinators worldwide. Numerous papers that provide evidence for decline have been increasing yearly as biologists and others begin to carefully examine the evidence and appropriate responses to the phenomenon. Various countries and organizations have responded by developing initiatives to address the wide range of issues surrounding the loss of pollinators. These range from focusing on new biological research to environmental education to political outreach. One of the issues frequently mentioned concerns restoring pollinators to areas that once supported them under natural or near natural conditions. In this paper, case history information on restoring native bee pollinators to disturbed tropical dry forest in Costa Rica is presented. The work is proving to be a challenge because of the necessity to develop considerable ecological information on bees and their new relationships to plants in disturbed environments. Finding appropriate monitoring methods for assessing trends in bee populations through time has also proved to be challenging. Finally, social and political issues, especially regarding bee habitat and land tenure, require attention that place biologists in new and unfamiliar outreach roles.

Domination of ground cover by a single invasive plant species does not provide a habitat inferior to a native cover mosaic in terms of invertebrate assemblages.

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Habitat disturbance to remnant melaleuca bushland at Coombabah Wetland Reserve has promoted the dominance of invasive plant species at ground level. Ground cover at the reserve comprises large stands very strongly dominated by either blady grass (*Imperata cyclindrica* var. *major*: Poaceae) or bracken fern (*Pteridium esculentum*: Dennstaedtiaceae) interspersed with smaller patches consisting of a mosaic of native plant species. Epigeal invertebrate communities were monitored within each of these three ground cover types using continuous pitfall trapping over 22 weeks to investigate quality of the habitat in terms of providing niches to support invertebrate species. Diversity assessment suggested that the habitats dominated by invasive plants were not inferior to native cover mosaic in terms of species abundance, richness and evenness. High levels of similarity (75% species shared) in terms of actual invertebrate species present, and the rank abundance of these species occurred within the three ground cover types. Ordination suggests that invertebrate community structure within native cover mosaic was the intermediate between the bracken and blady grass communities. Several species are blady grass specialists and a few are bracken specialists, whereas no species specialise in the native cover mosaic habitat. On the basis of this study, there is no evidence to support the theory that a greater level of plant ground cover diversity supports a superior invertebrate community in terms of either overall diversity or composition.

Resistance to multiple aphid species has been identified in a single *Medicago truncatula* cultivar: resistance involves both salicylic and jasmonic acid signaling pathways

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Aphids are serious pests of legumes and other crops worldwide. In contrast to plant-pathogen interactions, very little is known about molecular mechanisms underlying plant resistance to aphids. Australian breeders have introgressed *Acyrthosiphon kondoi* (bluegreen aphid) resistance into three popular cultivars and generated three pairs of resistant and susceptible near-isogenic lines in the model legume *M. truncatula*. Further characterization showed that each resistance line operates against a number of major legume aphid species. However, the magnitude of resistance varied depending on the *M. truncatula* line and/or aphid species. We have focused on one pair of near isogenic lines, A17 (susceptible) and Jester (resistant) in which a single dominant gene conditions resistance to bluegreen aphid. Jester also exhibits, to different degree, resistance to pea aphid, spotted alfalfa aphid and spotted clover aphid. Analysis of the transcriptional changes in defence related genes representing various signaling pathways suggest that both salicylic acid and jasmonic acid pathways are involved in the resistance of Jester to bluegreen aphid. We are currently taking high throughput approaches to analyse gene expression patterns during various compatible and incompatible aphid interactions in order to further understand this important plant-insect interaction.

Effect of chemistry of plant waxes on insect attachment

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Insect ability to attach to epicuticular waxes of plants must depend on the structural, mechanical, and chemical properties of the plant surfaces. In order to test the effect of the chemistry of the surface on insect attachment, we measured the surface energy (SE) of several selected compounds of plant waxes (C18-C22 hydrocarbons, primary alcohols, and n-alkanoic acids) and the traction force generated by ladybird beetles *Harmonia axyridis* (Coccinellidae) on these surfaces. Our results demonstrated that the water contact angle differs greatly between different surfaces; however, the total surface energy of selected substances slightly depends on the chain length and not on the presence of active groups. No significant difference in traction force was detected on different wax surfaces. This result can be explained by the ability of attachment pads to generate a lipid-containing secretion. Since total SE does not differ very much between surfaces, only it's polar and dispersion components vary, the nonpolar secretory fluid aids in generating strong attractive forces on surfaces with different contact angles of water.

How suitable is this climate for an invasive insect? Using a phenology model for landscape-wide assessments of risk of establishment.

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One of the basic requirements of an invading insect is that the climatic conditions in the invaded location satisfy its seasonal requirements. For a holometabolous, phytophagous insect invading a temperate location this implies that eggs will hatch coincidentally with the emergence of suitable host material, that the cold-hardy stage will coincide with winter, and that these events will coincide sufficiently each year for the continual survival of the population. A multi-generational phenology model can directly assess this criterion of the risk of establishment (ROE) by quantifying the likelihood that temperature regimes in the invaded location will consistently produce seasonal development. Landscape-wide estimates of the ROE require a geographically robust phenology model – one that produces equally reliable simulations of phenological development regardless of the location. A geographically robust model of European gypsy moth phenology has been used for a North American-wide assessment of the ROE. Modifications to the model to assess ROE of the Asian gypsy moth in New Zealand, and preliminary results will be described.

Modeling insect behaviour in a constrained universe: caterpillar foraging on 3D virtual plants.

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Models of insect-plant systems have traditionally been expressed using general-purpose programming languages, and more recently with plant modelling languages such as L-systems. Creation of special-purpose languages for expressing behavioural models that operate within a virtual plant environment will make the behavioural modelling job a lot easier. The basic requirements of such a system, such as expression of insect sensory capabilities, actions and "decision-making" processes, will be discussed, and a prototype language described. Individual-based models of caterpillars moving in a field of plants will be compared with models defined on a stochastic basis previously in the literature.

Decisions, decisions – host-plant assessment by adult generalist and specialist aphids with the same genotype

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Winged summer virginoparae (long-day, crowd induced) and the winged autumn gynoparae (short-day induced) are asexual forms of the black bean aphid, *Aphis fabae*. When reared from the same clone, they have identical genotypes, similar morphology but distinct behavioural and reproductive phenotypes. With regard to plant preference they differ in that the *generalist* summer form will accept and reproduce on many different host plants while the *specialist* autumn form preferentially accepts and reproduces on the spindle tree, *Euonymus europaeus*. A number of host-acceptance parameters were examined to distinguish between the decision-making mechanisms of these insects. Time from plant contact and from stylet penetration to the start of reproduction, time from plant contact and from stylet penetration duration prior to first phloem recognition, time from phloem recognition and number of phloem entries to phloem acceptance (>15 min access) as well as the incidences of phloem acceptance were assessed. One host common to both aphid forms, spindle, and four herbaceous hosts of the summer form, three of which they had not encountered previously, were used. It was hypothesised that the *specialist* autumn form would make more rapid, efficient decisions on plant suitability than would the *generalist* summer form. The results indicate that the decision criteria differ as much between the summer form on different hosts as between the summer and autumn forms on the single, common host.

Oviposition behavior of the Hessian fly and consequences for offspring growth and survival

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The Hessian fly-grass interaction is characterized by plant defense via *R* genes and insect adaptation via *avr* genes. Although Hessian fly adaptation to plant *R* genes occurs only during the larval stage, the ovipositing female initiates the plant-insect interaction. Hessian fly larvae can move within the plant but cannot move to another plant. The mated female has a relatively short time (ca. 4-6 hours) to find homes for the 200-300 eggs she carries and is constantly in motion during the oviposition phase, flying from plant to plant, examining leaf surfaces, and ovipositing eggs. During these behaviors, the female is exposed to a variety of visual, chemical and tactile stimuli. Based on these stimuli, as well as endogenous stimuli, the female makes decisions about her flight direction, and locations where landing, examining behaviors, and oviposition will occur. Many of the decisions made by the ovipositing female benefit the growth and survival of offspring, but some do not. I will discuss the constraints placed on the female by time, as well as her sensory systems and mobility. Oviposition behavior also will be discussed within the context of the co-adapted system of *R* gene plant defense and *avr* gene Hessian fly adaptation.

Host mediated impacts on the population structure of oak gall wasp communities

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Oak gall wasps (Hymenoptera: Cynipidae, Cynipini) are very selective about the oak section and species upon which they induce their galls for reasons that remain unclear. Here I present data on Iberian and central European oak gall wasp populations apparently structured by host oak species. The extent to which similar population structures exist in an obligate chalcid parasitoid is then examined to determine if this specificity is expressed in further trophic levels. The Palaearctic oak gall wasps *Andricus kollari* and *A. quercustozae* show a population divide just north of the Pyrenees mountains in southern France, marked by a switch in distribution of their sexual generation host from the eastern European Turkey oak *(Quercus cerris*) to the Iberian cork oak (*Q. suber*). This pattern is compatible with the existence of long-lived oak-specific ecotypes within each species, supporting the conclusion from existing interspecific analyses that oak shifts are extremely rare in gall wasps. In contrast, population structure of the parasitoid *Megastigmus stigmatizans* does not show a clear break in the same location suggesting that unlike their gall wasp hosts, these enemies are not oak specific. This finding has major implications for contrasts in the population structure of different trophic levels within oak gall communities.

Smart-autoinoculation system attributes to consider in using insects as vectors of mycoherbicides against weeds.

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Biological control of weeds has been traditionally centred on the use of insects and microbial pathogens as biocontrol agents. Each of these approaches has brought considerable success in weed control, but an understanding of the tripartite interaction between insect-weed host-pathogen might lead to synergism and further improve current control measures. Here, we propose the development of a smart-autoinoculation system based on a tripartite model, using insects as deliberate and enhanced vectors of plant pathogens as a novel weed control strategy. This approach proposes using pheromone-based systems to attract insects to the pathogen inoculum and thereafter infecting the weed ("lure-and-infect"). However, the potential for deliberately using insects to deliver plant pathogens to weed hosts could only be realized if a wide range of hypothetical tripartite system conditions is met. Each of the three components is ecologically and biologically complex, and understanding of these factors is crucial. This research will gather information to examine the feasibility of available system options i.e. full benefits and risks of these components. Hence, a strategic approach will be to examine the specificity of interactions between the insect, pathogen and weed. The objective of this paper is to identify a suitable model system(s) with minimal complexity and having components that are clearly defined.

Studying tritrophic interactions by manipulating the release of terpenoid volatiles with molecular techniques

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Parasitoids of herbivorous insects make use of odours emitted by herbivore-infested plants in order to locate their hosts. The odours are made up of blends of volatile organic compounds (VOCs) that show many similarities among plant families and can be roughly divided in "green leaf volatiles" (C6 aldehydes, alcohols and derivatives), terpenoids and aromatic substances. Maize (*Zea mays*) reacts strongly to *Spodoptera littoralis* caterpillar feeding. The resulting odour is highly attractive to females of the parasitic wasp *Cotesia marginiventris* but it remains unclear which are the key compounds mediating the attraction. The application of molecular techniques to this system has allowed modification of the pattern of released terpenoids by manipulating genes involved in terpenoid synthesis. In olfactometer assays we tested the attractiveness of maize knockout-mutants with reduced ability to produce specific terpenoid compounds and transgenic *Arabidopsis thaliana* plants expressing maize terpene synthases. The combination of molecular tools, analytical chemistry and behavioural studies provided new insights in the relative importance of terpenoid VOCs for the attraction of parasitoids.

Characterising mechanisms of resistance to defoliation in *Eucalyptus grandis*

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Leaf chewing beetles, mainly Chrysomelids (*Chrysophtharta cloelia* and *Paropis atomaria*) and Christmas beetles (*Anoplognathus* spp.), are major eucalypt defoliators and commonly cause significant defoliation of plantations of commercially important eucalypt species. The presence of eucalypt genotypes resistant to particular insect herbivores has been recognized for some time and chemical factors have been the focus of most studies (e.g. Christmas beetles). In this study State Forest genetic field trials of *Eucalyptus grandis* were assessed for canopy damage, predominantly derived from *Paropsis atomaria* activity, to identify open pollinated families (OPFs) resistant and susceptible to defoliation. Three selected resistant and susceptible OPFs and additional clonal resistant material were then tested via multiple bioassays to assess key parameters such as relative growth rate, consumption rate, mortality rate, efficiency of consumption, oviposition preference and adult feeding preference of leaf material. The results of these bioassays will determine whether consistent quantifiable differences exist in physical or chemical attributes between resistant and susceptible OPFs and whether these factors affect both larval performance and adult host selection conjointly. We also demonstrate if cross-resistance exists for *Anoplognathus spp.* and Chrysomelids in resistant *E. grandis* individuals.

Functional genomics identifies candidate virulence proteins of phytoplasmas, pathogens of insects and plants.

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Phytoplasmas belong to the Class Mollicutes and are non-culturable intra- and extracellular bacterial pathogens of insects and plants. Various computer algorithms were employed to mine the near complete genome sequence of phytoplasma strain AY-WB for genes encoding potential virulence factors. So far, 56 candidate virulence proteins were selected for functional analysis. To determine whether these proteins affect plants, corresponding genes were cloned into the binary potato virus X expression vector, pGR106, and expressed in tomato and *Nicotiana benthamiana* via *Agrobacterium*-mediated inoculation. This revealed 16 phytoplasma proteins that induced morphological aberrations in plants. Interestingly, five of the 16 proteins contain nuclear localization signals. Localization studies using yellow fluorescence protein fusions showed that two of the five proteins target plant cell nuclei. Transcripts of genes of these two proteins were detected in plants during AY-WB infection. To allow immunolocalization of these AY-WB proteins in the leafhopper vector and plant hosts, genes were expressed in *Escherichia coli* for subsequent antibody synthesis.

Relaxation of induced indirect defenses of acacias following exclusion of mammalian herbivores

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Many plants offer rewards to attract ants that provide indirect defense against herbivores. These rewards include nectar and swollen thorn domatia, and their production can be inducible. *Acacia drepanolobium* trees in East Africa house ant defenders that reduce feeding by goats and juvenile giraffes. Theory predicts that costly rewards should be relaxed if the risk of herbivory is reduced, although this has not been previously demonstrated. We compared the rewards offered by trees experimentally protected from mammalian browsing for seven years with those offered by unprotected trees. Protected trees produced 25% fewer nectaries per leaf and 25% fewer swollen thorns than trees in browsed plots. Relaxation of indirect defenses when trees are protected from herbivores is consistent with the dual hypotheses that inducible defenses are dynamic and that plants can save costs by relaxing these defenses when they are not needed. Among trees exposed to mammalian herbivores, trees that were browsed within roughly the past year provided similar rewards for ants as did trees exposed to herbivores but not browsed within the past year. This result suggests that relaxation of indirect defenses only occurs following more than one year of protection from mammalian herbivory.

Insects as vectors of plant diseases - an introduction

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Viruses are among the most devastating of organisms inflicting plant diseases. In many cases virus diseases become epidemic, progressing rapidly through a field and severely reducing harvestable yields. What is more, once the epidemic has begun, it is extremely difficult to control and can lead to plowing down the field and ceasing to grow that particular crop in that area in the future. The vast majority of plant-debilitating viruses are difficult to control because they are vectored by insects. This symposium examines interactions between viruses and their insect vectors as well as the feeding and movement behaviors of vectors in an attempt to uncover new ways to manage epidemics. Remarkable progress is being made in discovering, developing, and deploying such management tools.

Complex interactions between bottom-up and top-down control of grasshopper (Acrididae) performance

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Grasshopper species from North American high plains grasslands experience highly variable environments, especially with respect to climate and the nutritional landscapes that vary within and among years. Long-term studies of the most common grasshopper species indicate corresponding variability in grasshopper population abundances. Strong evidence of density-dependence suggests the importance of biotic influences on responses in addition to extrinsic climatic effects. I will discuss underlying interactions between 'bottom-up' (food plant resources) and 'top-down' (natural enemies) factors that affect individual grasshopper performance in *Ageneotettix deorum* (Scudder), a common grasshoppers in Nebraska sandhills prairie. Relationships between bottom-up and top-down forces to grasshopper populations are important, subtle and complex. I will summarize our understanding of these interactions to date, emphasizing the importance of host plant quality for understanding the impact of natural enemies. Key points include evidence for strong indirect, trait-mediated responses by grasshoppers in the presence of spiders, density-mediated compensatory effects between predation and food quality, and possible threshold responses affecting population densities among years.

Behavioural changes in lepidopteran larvae on crop plants.

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Helicoverpa armigera Hübner (Lepidoptera: Noctuidae) or heliothis is a highly polyphagous major pest of crops world wide. Larvae cause direct damage primarily to reproductive parts of plants and to leaves. The influence of plant architecture (stage and crop type) on foraging by this species is not well known. This is particularly true for early instar larvae which are the key stage targeted for control. We show how foraging behaviour (movement and feeding) changes with crop type. Two crops *Vigna radiata* (L.) R. Wilczek (Leguminosae) (mungbean) and *Cajanus cajan* (L.) Millisp. (Fabaceae) (pigeon pea) were compared at the seedling and flowering/early fruiting stage. Mungbeans are short, large leaved and hairy compared to the more slender glabrous pigeon pea. Experimental conditions reflected Australian summertime temperatures. The amount of time allocated to key activities such as feeding and searching were determined from observations of newly hatched neonate larvae on whole plants over 8 hours. In addition the frequency and duration of a single bout and where larvae spent most time on each crop type were recorded as were the location and type of plant part most frequently fed on. Results show differences occur between seedling and flowering plants and crop type.

How the clover root weevil finds white clover roots in the soil

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The clover root weevil, *Sitona lepidus*, is a destructive pest of white clover. The adult weevil feeds on leaves above-ground whereas the soil-dwelling larvae attack the root system. Larvae initially feed on root nodules that house symbiotic N_2 fixing Rhizobia bacteria, before moving onto progressively larger roots. Studying such small soil insects *in situ* is difficult, but the development of X-ray microtomography has allowed us to non-invasively track the movement of larvae towards roots in the soil. Using X-ray microtomography and other techniques, we have shown that newly hatched *S. lepidus* can recognise white clover roots in the soil from 6cm away and distinguish them from non-host plant roots such as subterranean, strawberry clover and perennial ryegrass. Using these empirical results, an individual-based lattice Boltzmann model has been developed to simulate the movement of *S. lepidus* in the rhizosphere of white clover.

Interspecific competition between the native and the exotic dandelion mediated by pollinators

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In Japan, especially in the suburbs, the native species of dandelion has been replaced by the exotic one for the past few decades. This phenomenon is called "Dandelion Wars". The widely accepted hypothesis to explain this replacement is, that there is a habitat segregation between the two, i.e., the native prefers stable while the exotic prefers destructive, labile environment, which is spreading in the suburbs, and that there is no interspecific competition between the two. Is this true ? At least, we often find mixed population in the field, which means that there is not a clear habitat segregation. Here we propose another hypothesis, that is, there must be some indirect interspecific competition mediated by pollinators in such mixed population. The results of this study supported this hypothesis in three-holds. 1. The exotic flowers attracted twice or more the flower visitors than did native ones in the field. 2. Seed-set of the native species is 10 – 20 % higher in the single-species population than in the mixed population. 3. (omitted). The above results showed for the first time that the pollination of the native dandelion is interfered by the exotic one as the latter spreads its habitat.

Host-specificity and evolution of the genus ***Bruchidius sl.*** **(Coleoptera: Bruchidae).**

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The understanding of the mechanisms explaining the diversification of phytophagous insects is a stimulating topic and numerous studies have followed one another since Erhlich and Raven's (1964) seminal paper was published. Within the superfamily Chrysomeloidea, the family Bruchidae exhibits a strong host-specificity that strengthen their usefulness in evolutionary studies. Seed sampling in the field and monitoring of adult emergences give the opportunity to identify insect-plant associations accurately. By using molecular phylogenetics, we have investigated the relation between host-plant preferences and bruchid phylogeny in order to highlight possible evolutionary patterns. We have studied European and African members of the genus *Bruchidius sl.* separately, with the inclusion of additional data on the nature of seed secondary compounds for the later. Our results indicate that phylogenetically related bruchids are generally associated with phylogenetically related host-plants. However, for European species, the insect-host plant pattern does not follow the host-plant phylogeny recently proposed by Wink & Mohamed (2003). Morevover, for African species, it appears that host-plants phylogeny alone cannot explain the observed patterns. Interestingly, the nature of the seed secondary compounds appears to be the major factor explaining the diversification of *Bruchidius sl.* that specialized on Caesalpinioideae and Mimosoideae superfamilies.

Development of an *in vitro* feeding system for root-feeding Hemiptera

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Limited published data exists on artificial diets for root-feeding Hemiptera. Using Grape Phylloxera, *Daktulosphaira vitifoliae* Fitch (Hemiptera: Phylloxeridae) as a model insect, an artificial feeding system was developed. Phylloxera is classified as viticulture's worst insect pest due to damage caused to the root system of the commercial grapevine, *Vitis vinifera* L. Phylloxera feeds on non-vascular cells, inducing the development of a 'nutrient sink' and root galls, leading to a decline in vine health and eventual death. The development of an artificial feeding system for phylloxera required special chamber design due to the size of the insect (1 mm adult length). Specific requirements included allowing for diet renewal without insect disturbance, and reducing condensation levels on the diet membrane while maintaining a sufficient humidity level. Limited probing activity and survival of 1st and 3rd instar insects has been recorded for up to eight days. Diets containing amino acids improve feeding rates due to the insects natural diet of the high protein content of non-vascular cells. Optimising an artificial diet for phylloxera will determine of the essential nutrients for survival and development, and provide a medium for the rapid testing of potential control agents. The system developed in this study could be modified for other root-feeding Hemiptera.

Genetic analysis of aphid resistance in a plant model system

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Medicago truncatula Gaertn. (barrel medic) is grown as forage in Australia, where plant breeders have identified aphid resistant *M. truncatula* germplasm. The status of *M. truncatula* as a model legume provides the opportunity to study defence against aphids at physiological, biochemical and molecular levels. Aphid resistance has been backcrossed into susceptible cultivars to create pairs of resistant and susceptible near-isogenic *M. truncatula* lines. We are focusing on the near-isogenic lines Jemalong (susceptible) and Jester (resistant) to identify mechanisms of defence against *Acyrthosiphon kondoi* Shinji (bluegreen aphid). We have identified three modes of resistance to *A. kondoi* in Jester: antixenosis (non-preference by the aphid); antibiosis (negative impact on aphid biology); and tolerance (little plant damage under aphid infestation). Molecular-genetic analysis of the trait is underway to isolate and characterize the dominant aphid resistance gene, *AKR* (*Acyrthosiphon kondoi* resistance). This gene is tightly flanked by resistance gene analogs of the NBS-LRR gene family, which confer resistance to a broad range of agricultural pests including microbial pathogens and phloem feeding insects. Our results suggest *AKR* may also be a member of this gene family. The advantages of studying this trait in a model system will be discussed.

Induction reduces performance of a specialist herbivore on one host plant species but not another

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Induced resistance to herbivory is often thought to be more effective against generalist and semi-specialist herbivores than specialists. However, we found that even for specialists, the effect of induced resistance on herbivore performance may depend on host plant species. Collard (*Brassica oleracae*) and mustard (*Brassica nigra*) were grown in outdoor chambers; half of the plants were mechanically damaged to simulate herbivory. Induced resistance in collard decreased the performance of *P. rapae. P. rapae* feeding on induced collard had more difficulty digesting leaf material and grew more slowly. Conversely, induced black mustard resistance did not affect the performance of *P. rapae*. *P. rapae* feeding on induced black mustard had greater conversion efficiency of food to biomass and lower consumption rates, but their growth rates were unaffected. In an effort to determine how host plant characteristics changed following damage, and what characteristics correlate with *P. rapae* performance, concentrations of sinigrin in black mustard and the relatively complex suite of seven common glucosinolates in collard were also measured. Glucosinolate concentrations will be discussed considering the proposed value of induction to increase the variability of the host plant.

Impact of Pea leaf and stipule morphological variation on Pea aphid predators

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It has been suggested that plants could be modified to enhance the activity of natural enemies of herbivores. Among the plant characteristics that could be modified are physical features such as plant morphology. We have been able to test hypotheses regarding the effect of plant morphology variation on the effectiveness and intraguild interactions of pea aphid predators by using near-isogenic lines of peas *Pisum sativum* L. These lines exhibit single gene mutations that drastically alter the shape of leaves and stipules. Increased leaf morphological complexity was found to be detrimental to the predation efficacy and plant surface search efficiency of *Coccinella septempunctata* L. adults and larvae. However, the efficacy of *Chrysoperla rufilabris* (Burmeister) larvae was unaffected by increases in complexity. This difference in predator responses was also observed in recent work on the effect of pea afila leaf and reduced stipule traits on the efficacy of these two predators. Predation by the chrysopid was enhanced only when afila leaf occurred together with reduced stipules. In addition to influencing the efficacy of predation on aphids, variation in plant morphology can legislate the occurrence of intraguild predation among the aforementioned predators. Results from work on this topic will be presented.

Induced resistance and susceptibility of brassica plants to diamondback moth

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Correlations between induction response and constitutive resistance in plants to herbivores have been speculated for a number of years. In laboratory and field experiments we compared the effects of exogenous jasmonate treatment to two closely related cabbage species on the host-searching and oviposition preference of the diamondback moth, *Plutella xylostella*. Jasmonate-treated Chinese cabbage (*Brassica campestris*) was less attractive than untreated Chinese cabbage to ovipositing *P. xylostella*, while jasmonate treatment of common cabbage (*B. oleracea*) made plants more attractive for oviposition by this insect. Similar effects were observed when plants of the two species were damaged by *P. xylostella* larvae. In the absence of insect-feeding, or jasmonate application, Chinese cabbage is much more attractive to *P. xylostella* than common cabbage. Inducible resistance therefore appears to occur in a more susceptible plant and induced susceptibility appears to occur in a more resistant plant, suggesting a possible balance mechanism between constitutive and inducible defences to a specialist herbivore. A further study, that uses a range of wild crucifer plant species exhibiting low through to high degrees of constitutive resistance, is underway to test this hypothesis.

European honeybees and Australian grevilleas: cross-cultural attraction

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European honeybees, like most pollinators, are expected to visit attractive plants more frequently, and consequently these plants may achieve greater fitness. Many Australian plants species are very attractive to honeybees (eg. large floral display), but facilitate pollination by birds and mammals (eg. Proteaceae). The floral structures of these plants mean that honeybee foraging behaviour may not transfer pollen. Thus, investment in reproductive effort comes at a high price. Few Australian studies have tested the response of honeybees to variation in plant reproductive effort and the consequences for reproductive output. *Grevillea macleayana* **was used to (1) identify interplant variation in honeybee foraging behaviour (2) determine whether variation in plant reproductive effort (eg. inflorescence production) explained the interplant variation in honeybee foraging and (3) determine whether interplant variation in reproductive output could be explained by variation in honeybee foraging. Honeybee visitation was observed at 16 plants, at three sites in Jervis Bay (NSW), over 6 days. Significant interplant variation was found in rates of honeybee visitation. Significant positive relationships were found between rates of honeybee visitation per plant and both inflorescence number and plant size. However, thus far, no relationship has been found between plant seed production and rates of honeybee visitation.**

Depauperate pollinator and seed disperser assemblages provide adequate ecological services to an entomophilous and myrmecochorous shrub in restored habitat

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Concern over the loss of natural ecosystems to urban development has triggered many restoration projects. However, the effectiveness of various techniques used to restore biodiversity and associated ecological functions remains largely unassessed. We evaluated the success of a restoration project by comparing pollination and seed dispersal in an entomophilous and myrmecochorous shrub *Dillwynia sieberi* in revegetated pasture and remnant Cumberland Plain Woodland, Sydney, Australia. Flower visitation data from revegetated areas suggests a negative interaction between introduced Honey Bee and native bee species on *D. sieberi* flowers. This interaction was not pronounced in forest remnants due to the lower flower densities. Plants in restored areas had higher flower densities, while forest remnant plants had a higher flower-to-fruit ratio. Rates of *D. sieberi* seed removal by ants in revegetated and forest areas were similar, although species richness of seed-removing ants in revegetated areas was lower and their composition was different from that in forest remnants. Our data suggests that, although habitat restoration can take decades, recovery of certain processes occurs at the early stages of restoration due to the ability of certain species to expand their role in the ecosystem.

Density dependent effects of aphids on defense expression in milkweeds and aphids

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Density effects of the specialized sucking herbivore, *Aphis nerii*, on cardenolide expression were measured in a milkweed host plant with high constitutive defence expression. We found that host cardenolides were reduced at low aphid density and as density increased cardenolides were induced to levels above aphid-free controls. The cardenolide response was strongest in leaves with aphids, but a similar, less pronounced response also occurred in adjacent leaves. We could find no effect of cardenolides on aphid performance and there was no effect of time of exposure or the interaction between herbivore density and feeding time. *A. nerii* sequesters cardenolides for defensive use against its natural enemies, and seems capable of effectively handling significant increases in cardenolide concentration with little adverse effect on colony density and biomass. Cardenolide reduction and induction host plants may therefore influence the effectiveness of sequestered chemical defense against natural enemies in the third trophic level according to herbivore density. We explore the predicted implications of such dynamic shifts in plant response for both herbivore and natural enemy trophic levels.

Captures of Mexican fruit fly in orange baited traps, in Yautepec, Morelos, México

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The Mexican fruit Fly is one of the most serious pests of fruit crops in México and many other tropical and subtropical regions. Historically, the principal capture system is integrated by McPhail traps baited with commercial protein, use of host volatiles to bait traps is recent. The objective of this research was to compare capture rates of Mexican fruit flies in traps baited with commercial protein bait (Captor 300®), and in traps baited with orange fruit. From early february to late march, field experiments were conducted in a 14-ha mango orchard. The following treatments were conducted (1) orange pulp, (2) orange skin, (3) orange with pulp and skin, and (4) commercial protein. There were 4 replicates of each treatment and the position of trap was rotated every week. All captured counts were made weekly, all specimens were preserved in 70% ethyl alcohol. Fruit fly capture data for each treatment were subjected to a repeated-measures ANOVA using weekly counts ad repeated measures, and treatment means were compared using the LSD test. Data were transformed by log (x+1) to stabilize variances before analysis. In field tests traps baited with orange complete were more effective for capturing fruit flies than others baited traps (proportion 1.68:1 versus commercial bait, 2.95:1 versus pulp alone, and 1:1.55 versus orange skin). Results from this study indicate that use fruit odors as attractants in commercial orchards will improve the number of flies captured.

Behaviour of first instar *Helicoverpa armigera* (Hübner) (Lepidoptera: Noctuidae): Does starting position make a difference?

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The effect of oviposition site on consequent behaviour of first instar *Helicoverpa armigera* (Hübner) was investigated on cotton plants. Larvae were observed from egghatch for three hours starting from three positions: upper leaf, square, and boll. Of the following behaviours quantified using Noldus observer: feeding, crawling, resting, dropoff (falling from the plant on a silk strand) feeding on the eggshell, waving down, waving up, and unknown, significant differences in the total amount of time spent feeding, crawling, resting and dropoff were found among treatments. Larvae that started from the upper leaf rested most and crawled least. Larvae that started from squares fed most and dropped off least. Larvae that started from bolls crawled most and fed and rested least. Further study is needed in order to discover the mechanism for the behavioural differences and to determine if the behaviour of larvae from different oviposition sites differs over longer time scales. Implications of oviposition sites on the foraging behaviour of first instar larvae and its relevance to the management of *H. armigera* in the field are discussed.

Pollination of a high latitude, dioecious plant, the cloudberry (*Rubus chamaemorus* L.): a generalized or specialized interaction?

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Recent articles on the latitudinal trends in plant-pollinator specialization have questioned the long held notion that proportionally fewer specialized interspecific interactions occur at higher latitudes. We conducted a study to examine the pollinator guild of cloudberry (*Rubus chamaemorus* L.), dioecious, perrenial herb in subarctic bogs. We recorded 43 different insect Families as visitors, suggesting generalization in this pollination system. yet the Apidae, Halictidae, Muscidae, and Syrphidae represented 87% of all visitors. Controlled pollination experiments revealed that the halictids and syrphid were the most effective pollinators although the mass and seed set of the fruit actually produced did not differ between the four Families. Based on pollinator importance, a product of relative abundance and effectiveness, showed syrphids and muscids were of paramount importance to cloudberry as these Diptera were physically excluded from accessing nutritional rewards from competing Ericaceae flowers and consequently obligate visitors to cloudberry. Thus, there is moderate pollinator specialization with cloudberry offering small but significant rewards to attract small Diptera.

The ecology of inquilinism in communally parasitic *Tamalia* gall aphids

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Tamalia aphids occupy galls on *Arctostaphylos* spp. shrubs in Western North America. A gall-inducing species, *Tamalia coweni*, exhibits facultatively communal behavior in that multiple female foundresses may share galls. A second species, *Tamalia inquilina*, obligately invades galls of *T. coweni*, singly or in groups, although no apparent agonistic interactions have been observed between the two species. Although total reproductive output of adult offspring is similar between galls containing *T. coweni* alone and galls with both species, the allocation of brood is significantly skewed toward that of the inquiline; hence, *T. inquilina* is a parasite of *T. coweni*. The presence of *T. inquilina* has no significant effect on survivorship of *T. coweni* in mixed-species galls. *Tamalia inquilina* can successfully reproduce in open galls abandoned by *T. coweni* and under these circumstances may be characterized as commensal rather than parasitic. To the author's knowledge, this is the first example of communal parasitism in galling aphids.

"Toxicity of pellitorine, an amide isolated from *Piper tuberculatum* Jacq. (Piperaceae) to the pollinator *Apis mellifera* L. (Hymenoptera: Apidae) "

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Pellitorine, an amide isolated from *Piper tuberculatum* (Piperaceae), has been studied as a biopesticide against agricultural pests in Brazil. In order to investigate their impact on non-target organisms, particularly pollinator insects, researchs were performed to evaluate the acute toxicity action of the compound on larvae and newly emerged adults of honeybee *Apis mellifera* by means of contact and ingestion bioassays. Workers in the larval and adult phase were separated in groups, which received pellitorine in different concentrations. The larvae were maintained in their own original cells, receiving feeding and normal care from the nurses. The adults were confined in wooden cages with screens, receiving artificial diet made up of sugar and water (1:1). The concentrations of 40, 200, 1000, 5000 and 25000 ng a.i./individual were obtained diluting pellitorine in 98% ethanol. LD_{10} values of 39.14, 36.16 and 13.79 ng a.i./insect were determined for larvae, for adults by ingestion and adults by contact, respectively. The honeybee larvae were shown to be highly susceptible to the amide pellitorine. The amide pellitorine was shown to be more active when its absorption took place via dermal contact (by topical application).

A dynamic brain is the key to become a tospovirus vector

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Although tospoviruses are transmitted only by thrips, it is not possible for an adult to acquire then transmit one of these plant diseases. An individual thrips can acquire a tospovirus only by feeding on an infected plant during its first or early second larval instar, the virus then moving from the gut lumen to the salivary glands from where it can subsequently be re-injected into a plant. Much of the research by tospovirus workers has concentrated on how the virus enters the gut cells, but the translocation of the virus through the insect body has been given rather less attention. Studies on the developmental morphology of the early instars of *Frankliniella occidentalis*, the western flower thrips, have demonstrated a well defined period during which there is a temporary association between the mid-gut, the visceral muscles and the salivary glands. This association occurs because during the first and early second instars the brain is displaced into the prothoracic region by the large cibarial muscles. With increasing body size, the brain moves into the head and the close association is lost between the salivary glands and gut wall. Subsequently, there is a strong input of virus into the malpighian tubules from the haemocoel.

Host plant use by polyphagous grasshopper and the defense traits of the plants related with it

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Most of herbivorous insect species are oligophagous although the benefits to expand the host plant range were reported. That suggests that polyphagous insects have more cost in host plant use than oligophagous ones do. The one of the causes is considered that they have to defeat various plant defenses of family-specific. When they feed on unrelated plant species with common defense traits, such feeding habits could contribute to less cost in host plant use than ones with uncommon. However, a few studies examined the use of host plants growing in the habitat and the defense traits with them. Because most of grasshoppers were polyphagous at an individual, they have the feeding habits and mechanisms for decreasing such cost. Host use pattern by nymphs of polyphagous grasshopper, *Parapodisma subastris* Huang was examined, and the results were as following:1. Out of 19 plant species growing in the habitat, the plants with tough leaves or dense trichome ranged over family, and showed higher intrinsic quality. 2. When nymphs fed on several lethal plant species, they improved their survivorship greatly. Above the characteristics of host plants and the feeding habits were useful to decrease cost of expanding host plant over families.

Integrity of pollinator communities in restored habitats in Mauritius.

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Island habitats normally show high degrees of endemism but are often affected negatively by exotic invaders and species extinctions. The aim of our study is to assess the management success for the restoration of a native, original plant community in terms of essential ecosystem services, such as pollination and seed dispersal. We use quantified interaction webs of the pollinator community to contrast the quantified network between plants and pollinators in restored and unrestored plant communities. Restoration is achieved by regular weeding of invasive, exotic plant species, resulting in higher disturbance levels than in unrestored habitats. We show that abundance and diversity patterns of the pollinator interactions differ between restored and unrestored plant communities and that the resulting seed sets of endemic plants are affected by management. Experimental field studies are used to validate the predictions of strong influences of exotic invasive plant species on the pollination services to endemic plant species.

Endophytic fungi and their impact of insect food webs.

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Many fungal microorganisms live in symbiosis with terrestrial grasses. They affect both the quantity and quality of resources and thus, the energy supply to invertebrate consumers at higher levels in the food chain. Plants infected by endophytic fungi are chemically protected against herbivore consumption. However, the influence of this fungus-plant association on multi-trophic interactions remains largely unexplored. We show effects of fungal endophytes on insect food webs that reflect limited energy transfer to consumers as a result of low plant quality, rather than productivity. We use quantified herbivore-parasitoid webs on endophyte-free and endophyte-infected grasses to show enhanced insect abundance at alternate trophic levels, higher rates of parasitism, and increased dominance by a few trophic links on uninfected plants. These results mirror predicted effects of elevated productivity on food-web dynamics. Experimental analysis of the underlying mechanism indicate life-history effects for different species of herbivorous aphids and negative effects of endophyte presence on aphidophagous insects.

"Role of pseudomonad induced resistance in tomato *Lycopersicon esculentum* against leafminer, *Liriomyza trifolii*"

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"Tomato is an important vegetable crop of India and it is frequently infested by the leafminer, *Liriomyza trifolii* resulting in yield loss. Insecticides offer less impact in its management and genetic resistance iS minimum in commercial cultivars. Induced resistance is found to be a source of pest reduction in crops like tomato. Hence, an attempt was made to induce the resistance in tomato using biological and chemical inducers. *Pseudomonas fluorescens* (strain Pf1) was used to treat the seeds, seedlings and was applied to soil and foliage. The incidence of *L. trifolii* was significantly reduced on plant basis (29.7%) and leaflet basis (19.9%), when compared to check (53.7 and 31.9% respectively). The induced plants showed non-preference for feeding and egg laying to female flies. Antibiosis was observed in larval growth and development. Egg to adult period was increased by 3.2d. The larval parasitism by *Chrysaonotomyia rexia* was 34.7% more on the induced plants. Leafminer infestation resulted in enhanced expression of the pathogenesis related (PR) proteins and defense enzymes *viz.,* peroxidase, polyphenol oxidase, phenylalanine ammonia lyase, 1-3 glucanase, proteinase inhibitors and chitinase. The defense chemicals *viz.,* total phenols, tannins and acylsugars were also high in the induced plants. Compatibility of induced resistance was also noticed with the neem products under field condition.
"

Neural processing underlying taste-mediated behaviour in locusts

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The decision an insect makes to accept or reject food is largely a consequence of the information it receives from its taste receptors. These taste receptors, or contact chemoreceptors, are distributed over the body and mouthparts. How then is the chemosensory information provided by these receptors encoded within the central nervous system? Recent advances in our understanding of taste perception in insects shows that there is a very close anatomical and physiological relationship between chemosensory and mechanosensory processing. The destinations of chemosensory neurons in the central nervous system is determined not by the type of chemical that they respond to, but rather to the spatial location of the chemosensory receptors on the leg. Our recent studies show that local interneurons within the thoracic nervous system are responsible for much of the processing of chemosensory information from contact chemoreceptors on a leg. Chemosensory coding by these interneurons appears to be for a single quality, that of aversiveness, which is a product of both chemical identity and concentration. Moreover, the strength of the chemosensory responses in interneurons and leg motor neurons is strongly correlated with the behavioural aversiveness of a chemical stimulus.

What good are syndromes? A review of pollination systems in New Zealand.

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The concept of pollination syndromes, originally designed to describe a combination of floral traits that frequently occur together in association with particular groups of pollinators, has recently been criticised. Determining the most effective pollen transfer vectors (either in quality or quantity) is context dependent and often fails to match predictions based on the syndrome concept. We review pollination systems in New Zealand to assess the usefulness of the syndrome concept. The low diversity in native and exotic pollinator assemblages, extremely high seasonal and daily microclimate variability, and easy access to pollen and nectar in most flower types in New Zealand is thought to promote generalist and mixed pollination systems. The majority of native insect pollinators in New Zealand (e.g., small bees and flies) do not manipulate flowers with precise or complex behaviours. Therefore the syndrome concept as a predictive tool has been considered less appropriate than for North Temperate and Tropical regions (e.g., large bee and hawkmoth). However, differential floral attraction, floral preferences, and temporal resource partitioning need to be considered. Used correctly, the syndrome concept still has an important role in understanding and predicting the nature of pollination systems in New Zealand.

Effects of trait variation and weather on herbivore survival and on the strength of trophic interactions in a food chain

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Classical theory in community ecology assumes that smaller-scale details such as individual traits can be abstracted and that community dynamics can be characterized in terms of net changes in population densities. I explore the effect of body size variation among herbivores on community dynamics using a computational model which consists of a three-level food chain. I illustrate that survival of herbivores is negatively correlated with variation in their body size and that predicting herbivore population dynamics requires incorporating frequency-dependence. At the community-level, however, it is the proportion of surviving herbivores rather than their population density that determines plant abundance, i.e., plant dynamics can be predicted from the initial body size distribution of herbivores in combination with their body size-survival curve. I then present empirical evidence, for the relationship between net food web effects and variation in herbivore body size, from an old-field community, consisting of spider predators, grasshopper herbivores, and grass and herb plants. I show qualitative congruence between results of the simulation and field experiments. Decreased grasshopper survival not only reduces the damage inflicted on plants, it weakens the strength of indirect effects of spiders on grass and herb plants. Grasshopper survival, however, is also sensitive to intra- and inter-annual weather variability. Small changes in the distribution or duration of rainfall events result in changes of up to several orders of magnitude in the mortality rate of grasshoppers. I thus conclude that in this system both body size distribution of grasshoppers and weather should be incorporated in a time-series model whose goal is to predict community dynamics.

The plant signal cis-jasmone - genetics to applied ecology

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In many cases, volatiles produced by plant defensive mechanisms during herbivory act directly to repel insect attack, and may have indirect effects by attracting predators and parasitoids of the pest insect. *cis*-Jasmone occurs as a component of flower volatiles, but can also be produced by damaged vegetative tissues. In an olfactometer bioassay, *cis*-jasmone was repellent to alatae of the grain aphid, *Sitobion avenae*. Methyl jasmonate, a catabolite of jasmonic acid biosynthesised from linolenic acid during herbivory, can induce defensive metabolism in various plants. The biosynthetically related *cis*-jasmone is also a catabolite of the stress-produced jasmonic acid, but had previously been considered only as a biological sink for the jasmonate pathway. However, bean plants, *Vicia faba*, exposed to *cis*-jasmone vapour were more attractive to the parasitoid *Aphidius ervi*. This was associated with a more persistent effect than found with methyl jasmonate, including induced production of (*E*)-ocimene, a parasitoid foraging cue. The molecular mechanism by which *cis*-jasmone influences plant gene expression is being studied in *Arabidopsis thaliana*, with about 30 genes upregulated. These studies will potentiate development of *cis*-jasmone as a plant activator for crop protection, as well as allowing "switching on" of other useful genes and traits. Already, in a series of small-plot experiments conducted over four years, *cis*-jasmone applications have been shown to reduce cereal aphid populations infesting wheat in the field.

Can molecular tools be used to improve proximate and adaptive understanding of tritrophic interactions?

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There are increasing opportunities to use molecular tools which can allow further understanding of the mechanisms involved in herbivore-induced plant signalling to higher trophic levels. However, this should not lead to further segregation between the reductionist and holistic approaches to tritrophic interaction research. The use of transgenic/mutant plants and genomic tools such as microarrays should allow mechanisms to be explored at the same time as answering questions about the ecological and evolutionary roles of herbivore-induced changes in plant chemistry. Arabidopsis is proving a useful model-plant for exploring tritrophic interactions involving both aphids and lepidopteran larvae and their parasitoids. Using GM and mutant Arabidopsis, the role of specific chemical/physical traits has been explored, including herbivore/parasitoid behavioural responses to "novel" secondary compounds. Other studies are focussing on wild populations of *Brassica oleracea* using genomic tools being developed for Arabidopsis. Two populations of *B. oleracea*, which differ in aliphatic glucosinolate composition, are being currently investigated at the genomic, chemical, behavioural and ecological level. This is an example of how molecular tools can help develop our proximate and adaptive understanding of herbivore-induced plant defence systems.

Aphid-plant interactions during stylet penetration of the apoplast and symplast

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Aphids alter plant physiology via mechanical stylet penetration, injection of gelling and watery saliva, removal of photosynthates, and transmission of plant viruses. Incorporating insect and plant in an electrical (EPG) circuit allows monitoring of stylet activities during these processes. The DC-EPG system uniquely differentiates intracellular (symplastic) and extracellular (apoplastic) stylet tip positions. Although the stylets follow an apoplastic pathway, the maxillae regularly puncture cells with important consequences for host selection and virus transmission. I will consider evidence for plant defences against aphids in both apoplast and symplast:

- Apoplastic defences. Stylet avancement may be restricted by mechanical properties of cell walls. Video recording of labial retraction is being used to investigate relationships between the rate of stylet advancement and different 'pathway' activities.
- Symplastic defences. Stylet puncture of a phloem sieve element may be followed by injection of watery saliva and uptake of phloem sap. Components of aphid saliva injected immediately after phloem puncture probably inhibit normal sieve tube responses to damage (particularly P-protein gellation) and enable sap uptake without phloem sealing. Comparisons of insect responses to nearly-isogenic resistant and susceptible plants suggest that resistance factors are expressed in the phloem and may involve modified sealing mechanisms.

Non-persistent virus transmission by aphids: new evidence for the ingestion-salivation hypothesis

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Approximately 75% of aphid-vectored viruses are transmitted in a non-persistent (non-circulative, stylet-borne) manner. The non-persistent transmission mechanism has therefore been the focus of extensive research. Electrical recording of stylet penetration activities (EPG technique) reveals that aphids show three successive intracellular activities (sub-phases II-1, II-2 and II-3) during maxillary puncture of the epidermal plasma membrane. Sub-phase II-3 is linked with virus acquisition and therefore represents uptake of virions to their retention site within the maxillary food canal. Sub-phase II-1 is associated with release of virions into an inoculated cell. The 'ingestion-salivation' hypothesis speculates that inoculation occurs via salivary release of virions from the common food-salivary duct at the tips of the maxillary stylets (Martin et al., 1997, *Journal of General Virology* 78: 2701-05). I will report recent EPG experiments using inoculation of circulatively-transmitted pea enation mosaic virus as a marker for intracellular salivation during epidermal cell punctures. The results confirm that inoculation of non-persistent viruses (sub-phase II-1) is associated with injection of saliva directly into the cytoplasm.

Gall-inducing species adaptive radiation, biogeography and biodiversity

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The local richness of gall-inducing insect faunas reaches a peak in Mediterranean latitudes, and their altitudinal equivalents in the tropics. The validity of this pattern and the mechanisms that result in this pattern are debated, just as the adaptive significance of the galling habit itself is controversial. Such discussion of patterns and mechanisms stimulates many new studies, with encouraging expansion of research in the tropics. Adaptive radiation of gall-inducing lineages appears to be opportunistic rather than coevolved or cospeciated with host plant taxa, with some galling families radiating into many adaptive zones represented by different plant resources. Radiation on herbs, shrubs and trees is influenced by plant architecture, although the specifics depend on the particular host plant and insect herbivore interaction. The large genera and the many sibling species of some gall-inducing taxa suggest rapid speciation and adaptive radiation without much genetic differentiation, with more new species complexes uncovered each year. Studies on gall-inducing taxa will continue to illuminate many areas of ecology and evolutionary biology, although much research is required to fully understand these intriguing insects.

Herbivory modifies the genetic structure of birch populations

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We studied the effects of selective herbivory on genotypic diversity in seedling populations of silver birch (*Betula pendula*), a light demanding pioneer tree species. The study covered the first two growing seasons, during which considerable thinning occurred within the experimental populations. Mortality resulted in decreased diversity compared to a control simulating random mortality. When insect and vole herbivory and nutrient enrichment treatments were compared, taking into account differences in mortality, vole herbivory resulted in increased diversity in the canopy layer. This is noteworthy, since the canopy layer includes the seedlings with best prospects of surviving in competition for light. The 2-yr natural insect exposure had no effect on diversity when mortality was accounted for, but cluster analysis showed that insect herbivory changed the genetic structure of the experimental populations.

The transmission of the bacterium *Xylella fastidiosa* by sharpshooters

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The vector transmission of the bacterial plant pathogen *Xylella fastidiosa* (*Xf*) is deceptively simple. Virtually any xylem sap-feeding insect can be a vector, with no required latent period and molting arresting vector transmission. The simplest model for transmission is that the feeding-acquired bacteria attach to the foregut, and some bacteria detach during later feeding on other plants. The precise location(s) within the foregut from which *Xf* is transmitted is unknown, but < 200 live cells of *Xf* per insect are needed for efficient transmission, suggesting that the transmission-critical area of the foregut is very small. Colonies of *Xf* can "carpet" the vector's precibarium, but this does not increase transmission efficiency. *Xf* cells attach to the foregut cuticle most often where fluid flow is most rapid (5 to over 50 cm/sec). *Xf* mutants of genes for fimbriae (attachment filaments) and cell-cell signaling lack normal surface-attachment abilities and vector transmissibility. Anomalies that are not explained by a simple attachment-detachment model are (i) lack of transmission by vectors that acquire large numbers of cultured *Xf* from in vitro suspensions in sterile xylem sap, and (ii) the lack of transmission by non-xylem specialist leafhoppers that acquire *Xf* from feeding on plants.

The possible role of aphids' cuticular proteins in binding potyviral helper component

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Transmission of potyviruses by aphids requires the presence of the viral *helper component* (HC) protein. Apparently, the HC serves as a bridge between virions and the aphid stylet. Potyviruses were seen attached within the stylet's food canal. The stylet is made of epicuticle and composed of cuticular proteins (CuPs) and waxes. Herein, we test the hypothesis that CuPs bind to the HC, and thus, indirectly, bind to the virions. CuPs are insoluble after hardening. Therefore, several chemicals were used to extract cuticular proteins from whole aphids: 8M Urea, 1% Calcofluor, 1% Congo red or 5% Sodium dodecyl sulfate (SDS). The extracted proteins were separated by electrophoresis, electroblotted onto a nitrocellulose paper and exposed to a recombinant HC tagged with 7 histidine residues. Bound HC was visualized using a commercial monoclonal antibody to the His-tag. This procedure allows determining that CuPs bands of 31 and 33 kDa that were extracted with either 1% Congo Red or 1% Calcofluor to react with the HC. This is the first report for an affinity between the potyviral HC and the aphid CuPs. A biologically inactive HC did not bind to extracted CuP. The significance of this finding and future work will be discussed.

Parthenium hysterophorus (Asteraceae): the host plant for the gall moth *Epiblema strenuana* (Lepidoptera: Tortricidae) and the gall weevil *Conotrachelus albocinereus* (Coleoptera: Curculionidae)

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The gall moth *Epiblema strenuana* and the gall weevil *Conotrachelus albocinereus* have been introduced into the state of Queensland (Australia) as an initiative to biologically control the invasive weed *Parthenium hysterophorus*. Both the moth and the weevil infest and induce galls on *P. hysterophorus*, but only the gall moth is established in the weeded natural areas. Both insects attack the shoot axes of *P. hysterophorus* and induce fusiform galls, although *C. albocinereus* usually restrict their attacks to the main stems and rarely occur on the terminal and lateral meristems. All developmental stages of *P. hysterophorus*, viz., rosette, pre-flowering, and flowering stages, respond to *E. strenuana* by developing galls, though of subtly different morphologies, whereas only the younger, viz., the rosette and pre-flowering stages of *P. hysterophorus* respond to *C. albocinereus* with galls. *E. strenuana* damages the growth point, phloem, and associated parenchyma tissue, and reduces plant vigour; on the other hand, *C. albocinereus* damages xylem tissue in particular, which results in stress arising out of water inundation and consequent anoxia.

"Does feeding by autumn gum moth larvae induce changes in *Eucalyptus globulus* leaf chemistry?"

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We empirically tested whether branch defoliation (artificial and larval) of 2-year-old *Eucalyptus globulus* Labill. altered the abundance of specific plant secondary metabolites immediately and 8 months after the cessation of larval feeding. The abundance of even carbon length foliar wax benzyl alkanoates and in particular benzyl n-tetracosanoate, was of foremost interest as high levels of these compounds have previously been reported to be associated with reduced *E. globulus* defoliation caused by autumn gum moth (*Mnesampela privata*) larvae. We found no evidence that benzyl n-tetracosanoate was induced by defoliation, consistent with its operation as a constitutive resistance mechanism. Other *E. globulus* plant secondary metabolites, including essential oils, polyphenolic groups and additional foliar wax compounds, were assayed and in all cases their abundance was not significantly altered by defoliation. However, the level of foliar tannins immediately after cessation of larval feeding did display a trend that suggested elevated levels as the result of defoliation, though this trend was not evident 8 months later, indicating that, if real, the response was a rapid and not a delayed induced response. The level of foliar tannins was also negatively correlated to both average larval survival and average percentage branch defoliation, suggesting that foliar tannins may operate as toxins to *M. privata* larval feeding.

Fiji disease virus levels in populations of the sugarcane planthopper - role of host-plant virus titre, percentage gall area and plant morphology

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Fiji disease virus (FDV) causes Fiji leaf gall (FLG), one of the most important diseases of sugarcane in Australia. The acquisition of FDV by its Australian vector, *Perkinsiella saccharicida,* varies with the sugarcane cultivar on which the vectors were raised. Vector populations raised on tolerant and resistant cultivars have a lower incidence of FDV infection compared to populations raised on a moderately susceptible cultivar. However, the features of the infected host plants in determining hopper acquisition levels remain unknown. Cultivars of known resistance to FLG were propagated using infected planting material. Virus free hoppers were allowed to lay eggs on each of them and the resulting populations of vectors were sampled and processed for FDV infection. Host plant virus titre was obtained with real time RT-PCR. Leaf areas and the areas infected with FLG galls were calculated using image analysis software.

Gustatory processing in the central nervous system of the locust

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Chemosensory sensilla on the legs of locusts contain four chemosensory and one mechanosensory neurone. These sensory neurones project to their local thoracic ganglion somatotopically, such that the location of the sensilla on the leg is preserved in the relative location of the neurones' arborisations in the central nervous system. Both mechanosensory and chemosensory neurones project to the same region of neuropil, and no modality or sensitivity specific segregation could be observed. A behavioural assay established that the probability of a locust withdrawing its leg from a droplet of a chemical solution applied to the hind leg increased with chemical concentration. All tested chemicals, even nutrients, could elicit withdrawal responses but critically the concentration at which different stimuli became effective at eliciting the behaviour varied over several orders of magnitude. Chemosensory neurones make monosynaptic connections onto the same spiking local interneurones that receive mechanosensory sensory inputs from the leg. The relative responses of local interneurones to these same solutions, as well as their outputs onto leg motorneurones, closely correlated with the probability of eliciting a withdrawal response. We suggest that chemosensory processing in the thoracic ganglia directly assesses a chemosensory quality, that of aversiveness, rather than individual chemical identities and concentrations.

The guilds of sawfly gallers on willow (*Salix* spp.) species

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We studied the guilds of galling sawflies (Hymenoptera: Tenthredinidae) living on 138 Eurasian willow species in order to investigate whether the numbers of different galler guilds found on individual willow species depend on various ecological characteristics of the hosts (growth form, mean height, habitat type, local patchiness, role in plant communities, geographical area of distribution, size of distribution area, section in taxonomy, and tendency to hybridise). A total of 308 galler species in nine guilds of galls were found. Leaf folders (*Phyllocolpa* and *Pontania* spp.) were found on 88, leaf gallers (*Pontania* and *Eupontania* spp.) on 176, and stem, petiole, or bud gallers (*Euura* spp.) on 44 willow species. The numbers of sawfly species per willow species correlated positively among galler guilds. The following ecological traits of the willows explained a significant amount of variation in the total number of guilds per host species: size of distribution area, geographical area of distribution, role in the community, local patchiness, growth form, mean height, and habitat type. The size of the distribution area explained ca. 40% of the variation in numbers of galler guilds per host species, which shows the overwhelming role of the size of the geographic range in explaining probabilities of colonisation by gallers.

Plant waxes, insect attachment and aphid predators

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Abstract: The surface of plants can greatly affect the ability of predators to locate and capture their prey. All aerial, terrestrial plant surfaces are covered in epicutlicular wax, which varies among, and within, plant species in composition, quantity and morphology. We worked with two cultivars of peas, one with a normal wax bloom, a thick crystalline layer of wax, and one with a severely reduced wax bloom. The reduced wax bloom cultivar supported lower pea aphid populations in the field. Pea aphids had the same intrinsic rate of increase on both pea phenotypes showing that the reduced wax peas were not by themselves resistant to pea aphids. In fieldwork using exclusion cages, we showed that the apparent resistance of reduced wax peas to aphids was due to the presence of predators. In the laboratory, we showed that *Hippodamia convergens*, the major predator of pea aphids in Idaho, was more efficient at capturing pea aphids on reduced wax peas than on normal wax peas due to the increased attachment of the predators to the surface of reduced wax peas. In addition, we studied the interaction of predators on plants with and without the wax bloom.

Environmental consequences of the introduction of *Bombus terrestris* (Linnaeus) (Hymenoptera: Apidae) in Chile.

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Three major environmental consequences of the introduction of *Bombus terrestris* have been reported: displacement of native bees, pollination of invasive exotic plants and the introduction of parasites. This work is focused on the first two consequences in Viña del Mar (V Región). To determine modifications on the foraging behavior of native bees by *B. terrestris*, the correlations in visiting frequencies to the *Psoralea glandulosa* (Fabaceae) were estimated. Visiting frequencies of native bees, *Apis mellifera* and *B. terrestris* to the invasive weed, *Galega officinalis* (Fabaceae), were measured and preliminary quantitative comparisons of pollen carrying were performed. A significant negative correlation of visiting frequencies of *B. terrestris* and native bees was obtained for one period of the day. On *G. officinalis* patterns of visitation were variable in time. *A. mellifera* was more frequent, although an interaction between day time and bee species was involved in the differences of visiting frequencies. Finally, *B. terrestris* carries twice the amount of pollen than *A. mellifera*. The results are discussed on the basis of bibliographical antecedents of *B. terrestris* introductions, and predictions of further experiments.

Pollination and pollinator mediated selection in Australian sexually deceptive orchids

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Sexually deceptive orchids mimic sex pheromones and appearance of female insects and thus attract only male insects which pollinate the flowers in an attempted mating. Male mating behavior is crucial for pollination and pollinator attraction is highly specific. 1) In *Chiloglottis trapeziformis*, one compound, named Chiloglottone, was identified to attract the males of the Thynnine wasp *Neozeleboria cryptoides* (Hymenoptera: Tiphiidae) as pollinators. To examine selection imposed on the orchids by pollinators, we used synthetic Chiloglottone on artificial dummies to study the male mate-choice behavior in the pollinator of *C. trapeziformis*. 2) When males of the pollinator were given the choice between two dummies of different sizes, they preferentially attempted to copulate with middle size dummies over small size dummies. 3) When given the choice between two dummies with different amounts of pheromone, males preferred the larger amount of pheromone. Larger amounts of pheromone generally attracted more males than smaller amounts. 4) Orchid flower labella, the mimic of a female body, were significantly longer and broader than female bodies. Flowers also produced on average ten times more Chiloglottone than females. Under pollination limitation, the evolution and maintenance of these exaggerated mating signals is likely to be mediated by the here demonstrated male pollinator behavior.

Negative effect of purple loosestrife and reed canary grass on the diversity of wetland plant and moth communities.

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Invasive plants have the potential to reduce the diversity of species in plant and animal communities. We examined the negative effect of two invasive wetland plants, purple loosestrife and reed canary grass, on the species richness and diversity of plant and moth communities within 24 wetland study sites in the Pacific Northwest. We hypothesized that as the cover of the invasive species increased, the diversity of the local plant and moth community would decrease. Increasing cover of purple loosestrife and reed canary grass was associated with reduction in the diversity of wetland plant communities irrespective of the diversity measure examined. Moth species richness was positively correlated with plant species richness. Wetland hydrology, soil characteristics, and topography were measured to control for potentially covarying and confounding influences on plant diversity. Temperature, ambient light, and surrounding land-use were measured to control for potentially covarying and confounding influences on moth sampling and diversity. None of these variables was significantly associated with invasive species abundance. This strengthens the conclusion that the invasive species are the cause of the decline in biotic diversity.

Sites of oviposition and initiation influence structure and associated community complexity in *Diplolepis* (Hymenoptera: Cynipidae) galls on wild roses

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Cynipid wasps of the genus *Diplolepis* are restricted to inducing galls on various organ systems of wild roses. Most species oviposit into unopened leaf buds or the tissues in close proximity to them. Leaf gallers such as *Diplolepis polita* and *D. ignota* deposit eggs on the surface of folded leaflets whereas stem gallers such as *D. nodulosa*, *D. triforma* and *D. spinosa* embed their eggs within the stem tissues between immature leaves. Unlike most stem gallers, *D. fusiformans* deposits its eggs onto the young, current year stems. Multi-chambered stem galls are more structurally complex than single-chambered leaf and stem galls. Inducers within single-chambered galls suffer greater mortality due to inquilines and parasitoids than do inducers of multi-chambered galls. Examination of gall structures and complexity of associated insect communities reveal patterns that help explain relationships among species of the *Diplolepis* complex.

Evolutionary relationships between swallowtail butterflies (tribe Troidini) and their pipevine host plants

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Butterflies of the tribe Troidini (Papilionidae: Papilionini) use exclusively host plants in the genus *Aristolochia* (Aristolochiaceae). Each Troidini species also has a preferred *Aristolochia* species, possibly due to the plant chemical compounds. This close association between Troidini/*Aristolochia* is an adequate model for insect-plant relationships and coevolutionary studies. Seeking to test the hypothesis of close evolutionary relationships between them, we have sequenced three genes (COI, COII, and EF-1α) of tropical Troidini, and two genes (matK and a non-coding region (*trn*L-*trn*F)) of the *Aristolochia* species used by those butterflies. The phylogenetic analyses were performed with PAUP, using Maximum Likelihood on the combined data (3285 nucleotides of butterflies and 2173 of plants). We did not find a perfect match between the phylogenies of butterflies and their host plants. Using data on the presence/absence of aristolochic acids in *Aristolochia* we were able to infer that independence of these compounds appeared twice in Troidini evolution. The sister species *Parides zacynthus/P. neophilus* (bootstrap = 100%), use species with and without those compounds, respectively. The next step is to identify which patterns have evolutionary origins and which are obtained by ecological interactions.

Will trichome-based resistance of *Lycopersicon* impair IPM?

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The Australian tomato (*Lycopersicon esculentum*) industry used pesticides to achieve production estimated at $225.3 million dollars in 2000-2001. Pesticides are still widely used in the industry and can contribute to the degradation of environmental, and human, health. The introduction of trichome-based host plant resistance of wild *Lycopersicon* species into the cultivated tomato has the potential to reduce the use of pesticides and the negative effects associated with their use. The effect of trichome-based host plant resistance on arthropod pests is well documented, though insufficient research has focussed on sub-lethal effects on natural enemies and compatibility with IPM. Natural enemies that are used, or have the potential to be used, in Australian tomato production were exposed to the wild species *L. pennellii, L. hirsutum* and *L. cheesmanii*, and mortality, entrapment and predation/parasitism recorded. Multiple regression analysis was used to determine whether trichomes played a role in mediating observed effects. The implications of trichome-based resistance on IPM will be discussed.

Locusts as model systems: from neurones to swarms to human obesity

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Over recent years the study of locusts has made important contributions to a diversity of areas within the field of insect-plant relationships – and more generally within the biological sciences. Some examples include contributions to the study of the neural bases of behaviour, phenotypic plasticity, nutrient balancing, learning, nutrient-allelochemical interactions, evolution of host plant range, optimal foraging and resource allocation theory, individual-based models of population dynamics, ecological stoichiometry and community ecology. State-space models of nutrition that arose from work on locusts have also provided fundamental breakthroughs in the study of human obesity and in the optimal design of diets for farmed animals. In this talk we survey a selection of these and set the scene for other speakers in the symposium.

Of mites and movement: the effect of plant canopy connectedness on biological control

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Successful biological control in ornamental crops in the UK relies on preventing the establishment of pests within the crop. This means that predators have to locate sparsely distributed prey within the crop. The structure of the plant canopy, in terms of both spatial complexity and connectedness will therefore have a major impact on the ability of a predator to locate prey, and this will influence the effectiveness of biological control. Impact of these factors on the ability of a randomly searching predator to locate prey has been investigated using virtual plant software that simulates a range of canopy structures. The results show that both connectedness and spatial complexity play important roles in determining the time to prey location, along with an effect of the number of plants in the canopy. The more connected the crop, the shorter the time to locate prey, but as crop canopy complexity increases, then the time to locate prey increases. This has important implications for the way in which biological control agents are introduced into the crop, and suggest that plant canopy architecture should be taken into consideration when developing biological control strategies.

Host plant-associated genetic differentiation in the snakeweed grasshopper, *Hesperotettix viridis* (Acrididae: Melanoplinae)

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Studies of herbivorous insects have played a major role in understanding how ecological divergence can facilitate genetic differentiation. In contrast to the majority of herbivorous insects, grasshoppers as a group are largely polyphagous. Due to the relative lack of intimate grasshopper-plant associations, grasshopper-plant systems have not played a large part in the study of host-associated genetic differentiation. The oligophagous grasshopper, *Hesperotettix viridis* (Thomas), is endemic to North America and feeds on composites (Asteraceae) within the tribe Astercae. Previous work has shown both preference and performance differences between *H. viridis* individuals feeding on either *Solidago mollis* or *Gutierrezia sarothrae.* We examined the genetic relationships among 38 *H. viridis* individuals feeding on these plants both in sympatry and allopatry using 222 AFLP markers. Neighbor-joining analysis resulted in two distinct host-associated clades with 71% bootstrap support for host–associated monophyly. Analyses of molecular variation (AMOVA) revealed significant genetic structuring with host plant accounting for 20% of the total genetic variance while locality accounted for 0%. Significant genetic differentiation was detected between *S. mollis*-feeders and *G. sarothrae*-feeders even when the two were present in sympatry. These results suggest that *H. viridis* as a species is comprised of at least two distinct host plant-associated lineages.

Gene expression in bean plants induced by spider mites

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When infested by herbivorous arthropods, plants start emitting a blend of volatiles called herbivore-induced plant volatiles (HIPV). One of the ecological functions of HIPV is to attract the carnivorous natual enemies of herbivores. In addition, HIPV can mediate interactions between plants, and between plants ans herbivores. HIPV are knwo to be specific in terms of plant species, herbivore species, developmental stage of herbivores etc., and such specificity can result in the attraction of carnivorous natural enemies of the herbivores. An intriguing question is how plants produce and emit such specific blends of HIPV. To date, several signaling pathways, such as jasmonic acid-, ethylene-, and salicylic acid-related signaling pathway are know to be involved in the production of HIPV. In addition, post-transcriptional regulation may be involved. In this paper, we use a system composed of two bean plants [*Phaseolus lunatus* (lima bean plant) and *Lotus japonicus* (a mdel regume plant)] infested by two spotted spider mites (*Tetranuchus urticae*) to investigate the above question.

The effects of self-pollination on plant defense against insects

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Self pollination is prevalent among plant species. Self pollination provides plants with reproductive assurance when pollinators are limited and provides a 50% gene transmission advantage. However, self pollination reduces genetic variation, increases the expression of deleterious alleles, and typically results in reduced fitness. This fitness reduction is called inbreeding depression. Understanding factors that increase or decrease the intensity of inbreeding depression is a major objective of plant evolutionary biology. We propose to investigate the effects of inbreeding on anti-herbivore defense in plants. Plants defend themselves through tolerance of and resistance to herbivory. We tested the hypothesis that inbreeding decreases tolerance in the yellow monkeyflower (*Mimulus guttatus*) and that plants compensate for this loss of tolerance by increasing resistance traits such as trichome production. To do this, we conducted experiments that quantified the tolerance and resistance of monkeyflowers to herbivores. Resistance and tolerance tradeoffs are thought to be widespread in plants, but few studies have demonstrated these trade-offs. This study found a significant effect of inbreeding on defense. This effect varied among families. There was also a significant negative correlation between defense traits and herbivore performance.

Flower Visitors of *Annona* spp. in Japan

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" Bees are commonly used for pollination of agricultural plants. However, for quite a few plants species, they are not available. Annonaceous fruit trees such as cherimoya and atemoya are included to these plants. We surveyed flower visitors of cherimoya and atemoya in Japan to find out potential candidates of pollination agents. The field survey was conducted from 1998 to 2003 at five study sites in Wakayama, Mie and Okinawa prefecture. We found more than 40 species of arthropods most of which were beetles. They were; *Cortinicara gibbosa* (Corticariidae), *Haptoncus luteolus, H. ocularis, Carpophilus marginallus* (Nitidulidae), *Mimemodes monstrosus* (Rhizophagidae), two unknown species of the family Stephylinidae, and others. In addition to beetles, thrips and true bugs were occasionally found. However, no bees or butterflies were found. Flower visitor communities were diverse at family level among the study sites. Total number of individuals were mostly the same for families Corticariidae, Ntitidulidae and Stephylinidae. Number of species was largest in Nitidulidae. The index of diversity H' varied from 0.83 to 1.77, and 1-D from 0.3 to 0.78. The index of similarity of communities, Morishita's C-ramda, ranged between 0.001 to 0.756. The flower visitor communities seemed to reflect communities of surrounding areas.
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Gene flow risk assessments: laboratory studies of the potential impact of host plant quality and an 'escaped' *Bt* gene from GM oilseed rape on *Pieris brassicae* and *Cotesia glomerata*.

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Gene flow from *B. napus* to its wild relative *B. rapa* is the best model system for studying transgene movement between GM and native Brassicas. To accurately assess the potential risks of this scenario, both the effect of plant hybridisation resulting in a change in host plant quality and the direct effect of an 'escaped' gene need to be addressed. Host plant quality can have significant effects on herbivores and their parasitoids. Using commercial oilseed rape *(Brassica napus)*, a wild *B. rapa* population and their hybrid, the biological and behavioural effects on *Pieris brassicae* and *Cotesia glomerata* of host plant variation, have been examined. The main findings indicate that herbivore development varies significantly on the three host plants. No significant effects of host plant were noted during studies of parasitoid biology. The effects resulting from a simulated escape of a *Bacillus thuringiensis (Bt)* toxin were also investigated using interspecies hybrids generated by a *Bt- B.napus* x *B.rapa* cross, as well as fully introgressed offspring. Results showed that the *Bt* gene expressed in these plants had a significant impact on the biology of both the herbivore and parasitoid when compared with control plants in which the gene was not expressed.

Crucifer specialists - clever manipulation of the major host plant defense

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Most known biochemical mechanisms used by insects to overcome plant chemical defenses involve the detoxification of secondary plant products. However, a two-component defense such as the glucosinolate-myrosinase system of cruciferous plants appears to offer additional targets for herbivore interference. In *Plutella xylostella* (diamondback moth, Lepidoptera: Plutellidae) a glucosinolate sulfatase (GSS) in the larval gut prevents the formation of toxic hydrolysis products. In contrast, larvae of *Pieris rapae* (cabbage white butterfly, Lepidoptera: Pieridae), another crucifer specialist, utilize a gut protein designated nitrile-specifier protein (NSP) to redirect the hydrolysis reaction towards the formation of nitriles instead of toxic isothiocyanates. GSS in *P. xylostella* and NSP in *P. rapae* appear to be the key biochemical counteradaptations that allow these insects to feed with impunity on plants containing glucosinolates and myrosinases. This finding sheds light on ecology and evolution of plant-insect interactions and suggests novel, highly selective pest management strategies.

Host plant location: correlating electrophysiology with behaviour

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To understand the mechanism by which the lepidopteran stemborers *Chilo partellus* and *Busseola fusca* are controlled by a "push-pull" strategy in cereal production, chemical signals (semiochemicals), released by the highly attractive trap crops Napier grass, *Pennisetum purpureum*, and blue thatching grass, *Hyparrhenia tamba*, and by maize and sorghum crops, were isolated from the air above the plants using entrainment into a porous polymer. The volatiles collected were analysed by gas chromatography (GC) coupled to electrophysiological preparations from the antennae of the stemborers. Active compounds were identified by GC-mass spectrometry. Significant qualitative and quantitative differences in production were observed between the host plant species and between day and night periods, and could be related to differential attraction of gravid stemborers. These studies are providing understanding of the roles of key semiochemicals in mediating host location by stemborers.

Feeding behavior of whitefly nymphs

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Electrical Penetration Graph (EPG) studies have identified several waveforms produced by nymphs of silverleaf whitefly, *Bemisia argentifolii* Bellows & Perring. One waveform, waveform C, is produced during the stylet pathway from the plant surface to the phloem sieve element. It is similar to the analogous waveform C of adult whiteflies. Three waveforms, J, H, and L are produced after the stylets penetrate a sieve element. Waveform J is the first waveform to appear after penetration of a sieve element; it lasts usually less than a minute and doesn't appear again. After waveform J, waveforms H and L alternate with each other over and over. Waveform H was correlated with honeydew production indicating that waveform H is produced during ingestion. Waveform L is probably correlated with salivation into the sieve element. The transparent nature of silverleaf whitefly nymphs allows observation of muscles operating the stylets, cibarial pump, and salivary pump. Contraction of these muscles was correlated with specific EPG waveform components by using a split-screen video technique where microscopic images of the muscle contractions were recorded on the same video tape with the EPG waveforms that were being simultaneously produced.

Contrasting plant-insect responses in wild and agricultural systems: Unintended influences of crop domestication

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Multi-trophic studies looking at expected and unexpected changes in plant-insect interactions have revealed both direct effects and indirect system effects. Direct effects from changes in plant resource allocation patterns have altered both plant responses to herbivory through alterations in plant tolerance to damage or decreased plasticity in response to herbivory. Enhanced tolerance to herbivory has been associated with increased allocation to non-reproductive structures in tomato and maize such that optimization of allocation patterns will depend on projected cropping system type and expected damage levels. Similarly, interactions between allocation patterns and changes in photosynthetic potential have interacted to shape the relationships between allocation to plant reproduction and tolerance. Unexpected negative effects of crop domestication on natural enemies via production of physical refugia or changes in semiochemical signatures have been demonstrated in both direct contrasts of wild and domesticated sunflowers or in high-yield strawberry phenotypes. Effects of domestication were dependent on system traits, e.g. nitrogen, such that the conclusions are contextually dependent. Failure to consider fully all three trophic levels in the domestication process appears to have resulted in the failure of native co-evolved natural enemies to regulate their native herbivore host.

Host plant volatiles for the strawberry blossom weevil

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The strawberry blossom weevil (*Anthonomus rubi*) may cause serious damage on strawberry crops (*Fragaria ananassa*) by cutting the stalk of the flower bud. In order to develop new protection methods for strawberry against insect damage this study aims at identifying plant produced volatiles, detected and used in host location by the strawberry blossom weevil. Plant volatiles of cut green plant parts were extracted on Porapak-Q and analysed by gas chromatography-mass spectrometry. The green leaf volatiles were dominated by hexanol- and hexenyl-type alcohols and esters. Various monoterpenes, sesquiterpenes and aromatic compounds were also identified. When analysing intact leaves by the use of headspace solid-phase microextraction coupled with GC-MS, mainly monoterpenes, sesquiterpenes, aldehydes and long-chain organic acids were detected. Changes in plant volatile, especially by the relese of sesquiterpenes were observed due to mechanical injury in strawberry leaves and flower buds. By the use of gas chromatography linked to electrophysiological recordings the insect antennae have been screened for sensitivity to plant volatiles. The experiments showed sensitivity of the antenna to 18 different compounds, identified as aliphatic ketones, aldehydes and esters, aromatic alcohols and mono- and sesquiterpenes. The behavioural effect to these compounds is now studied in laboratory assay and in the field.

Direct effect of frugivorous invertebrates on seed viability of host plants

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Frugivorous invertebrates, such as fruit flies, are often assumed to have a negative ecological impact on host plants, in part as a result of the economic loss they cause to cultivated crops. However, this lacks empirical testing. Germination trials were used to quantify the direct effect that *Bactrocera tryoni* (Froggatt) (Diptera: Tephritidae) has on seed production and viability of three host plants from the family *Solanaceae*. Tomato, capsicum and eggplant were selected as host plants to challenge the movement and proximity of larvae to seeds with their different spatial arrangements between seeds and flesh. Successful germination of infested and control fruits was compared for the proportion of the total crop of fruits infested and the proportion of individual fruits infested. Results suggest that *Bactrocera tryoni* are not detrimental to seed viability and do not reduce seed number in these hosts. Although the plants used in these trials do not have an evolutionary history with *B. tryoni*, the results suggest economic damage cannot be equated to reduced fitness in these hosts and evolutionary interactions between fruit flies and their larval host plants need to be reassessed.

Differences in species richness of galling aphids on *Pistacia*: selection of hosts or selection by hosts?

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Three species of *Pistacia* (Anacardiaceae) are common in Israel. *P. palaestina* and *P. atlantica* each host 6-9 host specific galling aphids (Homoptera; Pemphigidae: Fordinae). In contrast, *P. lentiscus* is colonized by only one species, *Aploneura lentisci*. Selection of hosts: The colonizing sexuparae do not distinguish between species of *Pistacia* and frequently land on the wrong host. The reddish color of the new foliage may distinguish the deciduous *P. palaestina* and *P. atlantica* from the evergreen *P. lentiscus* in the spring. Selection by hosts: Different response of the host plant to stimulation by fundatrices of different aphid species is possible. Resin in the phloem of *Pistacia* may be a barrier for the phloem-feeding Fordinae. *P. lentiscus* in particular produces commercially-exploitable quantities of resin. *A. lentisci* may have evolved a unique mechanism to exploit this host. Phylogeny. The phylogeny of the Fordinae suggests several host shifts during their evolution. None of these shifts involved *P. lentiscus*, which belongs taxonomically to a different group of *Pistacia*. It is possible that the difference in species richness is a phylogenetic trait.

Host range and life history strategy of *Asphondylia sphaera* Monzen (Diptera: Cecidomyiidae): utilization of *Rhus* species (Anacardiaceae) as momentary side hosts besides main annual hosts, *Ligustrum* species (Oleaceae)

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The majority of *Asphondylia sphaera* (Diptera: Cecidomyiidae) individuals are univoltine and oligophagous, inducing fruit galls on *Ligustrum obtusifolium*, *L. japonicum*, *L. lucidum*, and *L. ovalifolium* (Oleaceae) in Japan. However, some emergent early spring individuals have two generations a year, utilizing flower buds of *L. japonicum* for the first generation from May to June and fruit for the second from June to the following May. The current field surveys and DNA sequence data revealed that the emergent early spring individuals also utilized, for galling, the flower buds of *Rhus succedanea* and *R. sylvestris* (Anacardiaceae) from May to June. Adults emerged from the flower-bud galls on *Rhus* were considered to return to *Ligustrum* and to join the univoltine or bivoltine individuals of *A. sphaera* inhabiting fruit galls on *Ligustrum* spp. Thus, *A. sphaera* is regarded as a polyphagous species, galling on plant species belonging to different botanical families, Oleaceae and Anacardiaceae. However, the polyphagy of *A. sphaera* is neither seasonal host alternation as has been noted for other *Asphondylia* species nor simultaneous utilization of plural plant families throughout the year. *Asphondylia sphaera* exhibits 'momentary polyphagy', utilizing *Rhus* species only in early summer as momentary side host plants.

The face of furanocoumarin induction in wild parsnip

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Although most, if not all, plants possess some form of inducible defense, detailed knowledge of the causes and consequences of induction are known only for a few plants. Among those few is wild parsnip, in which genotoxic furanocoumarins are produced in all organs of the plant but are inducible only in a subset of them. Induction of furanocoumarins in leaves is particularly amenable to examination. A novel method of precisely mapping the spatial distribution of furanocoumarins in leaves reveals that furanocoumarin induction is limited to those injuries that disturb the integrity of furanocoumarin-bearing oil tubes. The consequences of furanocoumarin induction include benefits-deterrency and toxicity--as well as costs for synthesis and reduced photosynthesis and growth. The furanocoumarin defense is not effective against all folivores, but its mobilization appears to conform to classical optimal defense theory.

The conservation genetics of endangered mutualisms: increased genetic differentiation and reduced effective population sizes in specialist bees.

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Oligolectic bees collect pollen from one or a few closely related species of plants, while polylectic bees visit a variety of flowers. Because of their more restricted range of host plants, it may be expected that specialists exist in more isolated populations, with lower effective population sizes (Ne) than do generalists. We tested this hypothesis by examining levels of genetic variation in five phylogenetically independent oligolectic/polylectic species pairs. Within each pair, the oligolectic member had fewer polymorphic loci, lower allelic richness, and lower expected heterozygosity than its polylectic relative. The pattern of reduced genetic variation in specialists was statistically significant. We also tested the hypothesis of greater isolation of specialist populations by comparing levels of genetic differentiation in one species pair. Again, populations of the specialist species had significantly higher levels of genetic differentiation than those of the generalist over identical geographic distances. Our data support the hypothesis of greater isolation and lower Ne in specialist populations, making them more prone to extinction. We suggest that in instances where bee specialists are involved in mutually codependent relationships with their floral host, these mutualisms may be endangered for genetic and ecological reasons.

Insect feeding behavior as a marker to evaluate the compatibility of different elicitors of plant resistance

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This work focused on interactions involving systemic acquired resistance (SAR) and induced systemic resistance (ISR). Laboratory experiments were done to compare the effects of plant growth promoting rhizobacteria (PGPR), an elicitor of ISR, and ActigardÔ, an elicitor of SAR, and combinations on feeding behavior of cucumber beetles and aphids. Results of cage experiments with cucumber indicated that cucumber beetle feeding was significantly greater on ActigardÔ-treated plants than on untreated control plants. However, the combination of PGPR plus ActigardÔ resulted in significantly lower feeding damage than occurred with ActigardÔ alone; suggesting that simultaneous induction of ISR by PGPR mitigated the stimulatory effect of ActigardÔ. Additional experiments to compare PGPR and ActigardÔ treatment effects on aphid feeding behavior were conducted using electronic monitoring of insect feeding (EMIF) techniques. Results of the EMIF experiments indicated that aphids feeding on cucumber plants where SAR was elicited by ActigardÔ exhibited an increased number of stylet probes and spent less time in phloem than aphids on untreated plants. As in the cucumber experiments, simultaneous induction of ISR by PGPR (in the combination treatments) mitigated this effect.

Analysis of quantitative trait loci for stem borer resistance in a DH rice mapping population

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In order to better understand the genetic basis of rice stem borer [mainly *Chilo suppealis* (Walker)] resistance in rice, efforts to identify the quantitative trait loci (QTL) related to the stem borer resistance have been done. The phenotype trials of resistance to the stem borer were tested in 1 subtropical and 2 tropical environments (totally three seasons). We mapped the stem borer resistance in a doubled-haploid rice (*Oryza sativa* L.) population of 154 lines [CT9993-5-10-1-M (CT9993)/IR62266-42-6-2 (IR62266)]. A genetic linkage map with 315 DNA marker loci was applied. A mixed linear model approach was used to detect QTLs with main effects, QTLs involved in digenic epistatic interactions and QTL by environment interactions (QEs). Several main-effect quantitative trait loci (QTLs) that related to rice stem borer damage traits (dead heart (DH) and white head (WH)) were identified. Among them loci located in intervals G187 – ME2_11 - EM14_1 - RZ997 in Chromosome 8, RM219 - RG553 - EM14_6 in Chromosome 9, C813 – EM11_11 – RG109 – ME10_14 in Chromosome 1 were repeatedly identified main effect QTLs. Each trait normally has several pairs of epistatic loci, QEs, and additive * epistatic * environment interactions. These results are helpful for fine mapping the stem borer resistance QTLs in rice and marker assisted selection in the future.

EPG evidence for local suppression of induced resistance by aphids

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Paper not available at time of production.

Epidemiology of glassy-winged sharpshooter-vectored Pierce’s disease: application to disease management.

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Paper not available at time of production.

The Entry of Whiteflies into Greenhouses – Distribution in Time and Space.

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We determined the distribution of the sweet potato whitefly, *Bemisia tabaci*, outside and inside a greenhouse (inside area 1000m^2) equipped with side windows covered with 50-mesh screens and four open roof vents. The study was in the Northern Negev region of Israel during the fall season of three years. Outside the greenhouse significantly higher numbers of whiteflies were trapped on the down-wind side. Both outside and inside, 70% of the whiteflies were caught between 7:00 to 10:00 and 25% were caught between 10:00 to 13:00. Inside the greenhouse, most whiteflies were caught under the roof vents. The number of whiteflies caught under the roof vents and the number of tomato plants infected with TYLCV were significantly higher under roof vents at the down-wind side. Under our experimental conditions air was usually sucked into the greenhouse through vent located down-wind and exhausted through the vent located up-wind. Our results indicate that roof vents were the major port of entry of whiteflies in this greenhouse. It appears that the pattern of air movement through the roof vents determined the spatial distribution of the whiteflies in the greenhouse. This information may be used to reduce the risk of whiteflies entry and establishment.

Pollinator-mediated effects of leaf damage on plant fitness

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The effects of herbivores on non-seedling plants are almost always non-lethal, but can still have large impacts on plant fitness via both direct and indirect pathways. When plants are attacked by herbivores, a cascade of responses typically ensues. Common plant reactions to foliar damage include altered allocation to root: shoot biomass, short-term increases in photosynthetic rate, and the induction of costly structural and chemical defenses in plant tissues. Less well documented, but perhaps more prevalent than originally thought, are changes in allocation to reproductive structures like floral size and rewards after damage. Moreover, induced chemical defenses are often systemic and may enter into floral traits and rewards. Changes in plant morphology and chemistry after damage, in turn, can modify the responses of pollinators to damaged plants. The ability of plants to recover seed production or to export pollen after damage may be hampered indirectly by pollinator limitation, as well as directly by resource limitation. In this chapter, I will review the literature and discuss some of my own experiments that point to pollinator-mediated effects of leaf damage on plant fitness. Such trait-mediated indirect effects of damage may be important sources of fitness costs to plants.

How aphid colonization indirectly affects early-and late-emerging herbivorous insects on the tall goldenrod

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We investigated insect communities on a perennial forb *Solidago altissima* in Japan. The most dominant species was an aphid *Uroleucon nigrotuberculatum,* which mainly occurred from June to August. The aphid was tended by an ant *Formica japonica* for honeydew. Moreover, the aphid colonization induced rapid branching and increased the production of new leaves in October. Therefore, we hypothesized that the aphid colonization indirectly affected not only co-occurring herbivorous insects through removal behavior by the attending ants, but also temporally-separated insects in autumn. To test this hypothesis, we conducted an aphid exclusion experiment. The aphids negatively affected the abundance of caterpillars and leafhoppers by the excluding behavior of the attending ants in early season. On the other hand, the aphids also affected the abundance of temporally-separated insects, such as scale insects and grasshoppers, in late season. Prior sucking by the aphids decreased the density of scale insects. The decreased scale insects may be due to changes in plant quality by the feeding of aphids in early season. The decreased density of scale insects resulted in a reduction of density of the attending ants to scale insects. Therefore, the density of grasshoppers increased because of the low impacts of tending ants.

Ecological stress of aldicarb on soil arthropods associated with coffee plantations.

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Soil arthropods have been considered by various authors as good indicators of environmental effect of pesticides. This study aimed to evaluate the impact of a systemic insecticide (aldicarb) on soil arthropods associated with coffee plantations. The experiment encompassed three treatments (control, 1 application – 10 g of Temik 150 (aldicarb)/plant applied in 12/16/1999 and two applications - 10 g/plant applied in 12/16/1999 and 10g/plant applied in 03/01/2000) and four blocks, evaluated once before insecticide application and at 13 different days after the insecticide application. It was measured the soil arthropod densities found in berleze traps and the data was submitted to multivariate techniques. The days of evaluation were considered as treatments replicates through time and the generated data set was analyzed by canonical correlation and redundancy analyses and multivariate repeated analysis. Aldicarb effect on the assemblage of soil arthropods was significant and the main group impacted was Acarinae: Galuminidae. As the variation on the abundance of the coffee soil arthropods community could not be explained by the variation in weather conditions, it is suggested that the insecticide was the major factor causing the effects described previously.

Two genetically distinct lines co-exist in populations of the endoparasitoid *Venturia canescens* **.**

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In a laboratory colony of the solitary endoparasitoid *Venturia canescens* (Hymenoptera: Ichneumonidae) two genetically distinct lines (named RP and RM) appear to coexist sympatrically. The two lines differ in a cluster of phenotypic characters, including ovarian morphology, parasitism behaviour and reproductive success. Controlled oviposition experiments showed that RM females produce fewer offspring under single egg parasitism but more under con-specific superparasitism, compared to RP wasps. Investigations of interlarval combat under *in vitro* conditions showed that the higher reproductive success of the RM line under conspecific superparasitism is due to a physiological difference between the newly hatched larvae of the two lines which results in an advantage to the RM larva independent of the order or time interval between ovipositions. Examination of field-derived wasps has found that wasps displaying the two phenotypic clusters occur in natural populations. A model, using an iterative approach with experimental data to predict the stable composition in a mixed population of wasps displaying RM and RP, suggests that the two lines will coexist sympatrically when overall rates of superparasitism are greater than 5 to 10%.

Effects of understorey diversity in pine plantations on pest Lepidoptera and their natural enemies

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Biodiversity in plantation forests is becoming more important to forest managers as government policy and foreign markets increasingly demand evidence of sustainable management of these forests. Protecting indigenous biodiversity not only assists in conservation efforts, but may also play a role in ecosystem functioning, leading to a reduction in the risk of pest outbreaks. In this study we investigate the effects of indigenous and exotic floral diversity in the understorey of a pine plantation in Canterbury, New Zealand, on pest Lepidoptera and their predators. Of particular interest is the native geometrid *Pseudocoremia suavis,* which has caused major defoliation in pine plantations in the past, and carabid beetles, which may prey on moth pupae. The data suggest that moths are more abundant where the understorey is less diverse, and that carabid diversity in pitfall traps is greater with increased understorey diversity. These results indicate a benefit of increased understorey diversity in plantations

Life cycles of some ground beetles (Coleoptera: Carabidae) in Kuban-Azov lowland (South Russia).

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We studied seasonal dynamics, spatial distribution, reproductive cycles of five widespread (*Harpalus (*s. str.*) rufipes* Deg., *H. tardus* Pz., *Brachinus elegans, Br. psophia* Dej., *Br. explodens* (Duft.)) and one endemic (*Carabus exaratus* Quens.) species of ground beetles in agrolandscape of Kuban-Azov lowland. Life cycles of *Br. elegans, Br. psophia* and *Br. explodens* are monovoltine (one year with overwintering imago). Life cycle of *C. exaratus* is supposed to be similar to those of the former species, however it also includes overwintering larvae of last ages. In this area, the life cycles of *H. rufipes* and *H. tardus* are realized as annual with two variants: summer reproduction and overwintering imago and summer-autumn reproduction and overwintering larvae. The life cycles of the later two species are supposed to be the more flexible in relation to environmental factors.

RECULTIVATION OF POST MINING AREAS WITH *QUERCUS RUBRA* – THE SUCCESSION OF GROUND BEETLE COMMUNITIES

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In the Lusatian mining area situated in the East of Germany, lignite mining has been practiced for nearly 150 years. An area of more than 80.000 ha, has been utilized so far for open cast mining. The investigated sites are found on acidic lignite rich tertiary sands characterized by extreme abiotic soil conditions (low soil pH). After dumping, amelioration and fertilisation, a large amount of spoils were claimed by afforestation. According to the extreme soil conditions, various tree species were put to the test for afforestation. Only little is known about the ground beetle communities of afforestations of alien plant species stands such as Red Oak (Kolbe 1992). In 1994, Red Oak stands (*Quercus rubra*) formed nearly 12 % of the recent tree distribution on post mining landscapes in the Lusatian mining area. The succession of ground beetle communities of afforested and Red Oak stands in the Lusatian mining area was studied using the chronosequence method. Mainly grass dominated (*Calamagrostis*, *Festuca*, *Poa*) afforestations are colonized in high species numbers and abundances. The species composition of the afforestations was mainly influenced by the species compositions of the surrounding areas. Xerobiontic to moderate xerophilous, heliophilous openland species, like *Calathus erratus* and *C. ambiguus* or *C. fuscipes* are dominating. With increasing age of the stands the ground beetle communities changed from typical openland communities to woodland communities. Species composition in the 20 to 30 year old Red Oak stands differs from those in afforestations. In these Red Oak stands are to be found the lowest activity densities and biomasses of the chronosequence. This corresponds to results of Kielhorn & Keplin (1999) and Vogel & Dunger (1991), carried out in studies on ground beetle communities of the Lusatian mining area. Possibly the combination of extreme site conditions (dryness, low pH of soil) with a very close canopy of Red Oaks (85 - 95 % of this strata), very reduced plant diversity of herbs, and very reduced cover of herbs (1 - 5 % of this strata) are difficult for colonisation of phytophagous insects and therefore also for the mainly zoophagous ground beetles. The species composition of nearly 40 year old Red Oak stands on reclaimed soils in comparison to the reference stands (ALT) differ in habitat and shade preferences, but is in accordance with the humidity preferences of a Oak stand on the reference sites. In the older stands of the chronosequence and at the reference sites numerous large ground beetles like *Pterostichus niger* and *Carabus spec.* were found to be dominant. Dunger (1997) and Wanner & Dunger (2002) found for some groups underdeveloped soil arthropod communities on reclaimed sites on the pyrite rich Lower Lusatian spoils low. The quantitative aspect is remarkable, especially in comparison to the Upper Lusatian Area. The body length of the ground beetles is also an important factor for preying, and it is adapted to size of preferred prey.Some small species with low dispersal abilities have not been found on the sites on reclaimed soils until now. Possibly they have not yet colonised these sites. *Dyschirius globosus* for example has a small dispersal ability. We found this species in Red Oak stands on at disturbed sites, but it is still missing in the chronosequence stands on former mining sites. Within 40 years after reclaiming and planting, we found ground beetle communities which are, according to species numbers and humidity preferences, comparable to communities on Red Oak stands on undisturbed soils. Activity densities and biomasses of ground beetle communities are after 40 years of colonisation lower than activity densities and biomass on undisturbed soils. There are also still differences within the shade preferences and main habitat types between communities on reclaimed and not disturbed land.

Predicting the potential distribution of *Sirex noctilio* (Hymenoptera: Siricidae), a significant exotic pest of *Pinus* plantations.

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Sirex woodwasp (*Sirex noctilio*) is not considered an important pest in its native range, however, where it has become established in the Southern Hemisphere it has become a significant pest of exotic *Pinus* plantations. The potential distribution of *S. noctilio* in Australia, South America, Africa, North America and China was assessed from a study of the insect's potential distribution and host range using the climate-matching program CLIMEX. *Sirex noctilio* is predicted to be able to establish in the majority of commercial *Pinus* plantations in Australia, South America (Uruguay, Brazil, Argentina, Chile, Paraguay) and Africa (South Africa, Zimbabwe, Tanzania, Uganda and Ethiopa) over the next 25 years. In China, *S. noctilio* is predicted to be able to persist in many areas where large-scale afforestation of susceptible hosts has occurred and is planned. Restriction of movement of infested logs and the Sirex Management Strategy will help reduce the impact of *S. noctilio* in Australia, South America, Africa and China. *Sirex noctilio* is predicted to be able to persist in many areas in North America, where large tracts of land are occupied by susceptible hosts in native forests. Exclusion of *S. noctilio* is the only feasible approach in Northern America.

Bracon vulgaris Ashmead (Hymenoptera:Braconidae) parasitism avaliation in two pests in different parts of cotton plants in Ilha Solteira, São Paulo, Brazil.

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The objective of this work was to investigate ecological aspects of *Bracon vulgaris* Ashmead (Hymenoptera: Braconidae) an ectoparasite of both *Anthonomus grandis* Boheman, 1843 (Coleoptera: Curculionidae), the boll weevil, and Pectinophora *gossypiela* Saunders, 1843 (Lepidoptera: Gelechiidae), the pink bollworm, key pests of cotton culture, in several countries of the world, including Brazil. The investigations were conducted in 1996 and 1997 in an untreated area at the UNESP Farm in Ilha Solteira (São Paulo state). The samplings were made of weekly collections of reproductive structures of the cotton (cotton squares, flowers and cotton bolls), from the third superior, medium and inferior parts. These structures were examined using stereoscope in the laboratory for the presence of eggs, larvae, pupae or adults of the parasite. Parasite population fluctuated with the host population with tendency to increase in parasitism from the middle to the end of the crop cycle reaching a maximum of 61.5% in boll weevil larvae in the superior third and 22,2% in pink boll worm larvae in the medium third. The parasite, demonstrated to possess great conditions for contributing to Integrated Pest Control Management in cotton allowing for preservation of the natural environment.

Life Tables of the Parasitoid ***Aphidius gifuensis*** **Ashmead and its Host** ***Myzus persicae*** **(Sulzer)**

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The life history data for both the parasitoid *Aphidius gifuensis* Ashmead and its host *Myzus persicae* (Sulzer) were collected in the laboratory. To take both sexes and the variable developmental rate among individuals into consideration, the raw data were analysed based on the age-stage, two-sex life table. The intrinsic rate of increase for *A. gifuensis* is 0.246/day. The net reproductive rate is 46.1 offspring and the mean generation time is 15.6 days. To incorporate the parasitism rate with the two-sex life table, the daily parasitism rate of *A. gifuensis* was collected for individual female. The mean parasitism rate is 92.3 aphids/female. The intrinsic rate of increase for *M. persicae* is 0.233/day. The net reproductive rate is 29.4 offspring and the mean generation time is 14.5 days. The net parasitism rate for *A. gifuensis* on *M. persicae* is 46.2 aphids.

Spatial heterogeneity affects spider assemblages in a desert agroecosystem

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Desert agroecosystems are characterized by extreme differences in productivity, microclimate and habitat structure between the managed fields and the surrounding natural habitats. These differences may affect crop colonization by potential natural enemies such as spiders. We collected spiders from winter wheat fields and the surrounding natural habitats in the Negev desert in southern Israel and used pitfall traps, emergence traps and visual searching to determine species composition and relative abundance of spiders in this agroecosystem. Data show that there are three dominant families in the fields during the growing season: Linyphiidae are residents in the fields, and Gnaphosidae and Theridiidae are migrants from surrounding natural habitats and must colonize the crops each growing season. Linyphiide spiders survive in the fallow fields between successive crops. Indeed, only species belong to this family complete a full life cycle, from eggs to reproducing adults, during a single growing season. Implications for the role of spiders as natural enemies in desert agroecosystems are discussed.

Population variability of leaf beetles in managed and natural willow stands

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It is generally believed that insect outbreaks more frequently occur in managed systems, such as forest plantations and crop fields, than in natural and diverse systems. In this study we compare fluctuations in leaf beetle (*Phratora vulgatissima*) population density between willow (*Salix viminalis*) plantations and natural willow (*Salix cinerea*) stands. The primary aim was to test the diversity-stability hypothesis, assuming that plantations are less diverse than natural stands. Density of leaf beetles was measured in 20 willow plantations and in 22 natural willow stands for five years. Densities of main predators (e.g. omnivorous heteropterans) and two habitat diversity indices (plant species diversity and stand structural complexity) were estimated. Leaf beetle populations were, on average, as variable in managed willow plantations as in natural willow stands. Neither were there any significant relationships between habitat diversity and population stability of leaf beetles. The highest variability was found in some of the willow plantations where the leaf beetle went from relatively low density to high outbreak density within a few years. This pattern was not observed in any of the natural willow stands. Leaf beetle outbreaks were correlated with a low abundance of predators.

Temporal-spatial patterns of beetles and planthoppers across grass-heather mosaics in upland Scotland

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Grazing herbivores alter plant species composition and structure in upland indigenous biotopes. They do not uniformly forage and will typically select sheltered areas, feed in preferred patches and on favoured plant species within these. We are investigating how insects respond to the spatial heterogeneity of vegetation mosaics generated by domesticated sheep or wild red deer. We hypothesised that the size and structure of grass and heather patches would alter the distribution of insect species by affecting their rates of movement and ability to locate certain patches in the mosaic. We studied the distribution of small planthopper species, during their sedentary nymphal stage, on transects positioned across patch boundaries. The movement of large predatory ground beetles was recorded using a harmonic radar tracking system. The distribution of planthopper nymphs closely reflected the patterns of host plants, whether grasses or shrub species. The sinuosity of movement of predatory beetles did not differ between individual grass or heather patches but was significantly different across grass-heather mosaics of different grazing history. A trade-off was apparent for patch use by beetles between structural complexity and predation risk. We interpret these results to provide insight into the conservation of general insect diversity with grazing management.

Modelling the population dynamics of *Helicoverpa armigera* (Hübner) in Punjab, India

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A simple population dynamics model was developed to predict the incidence of *Helicoverpa armigera* (Hübner) on cotton using DYMEX (CSIRO, 1999). It is a computer package that enables interactive modelling of fluctuating populations of organisms in changing environments and has been used to model the population dynamics of *H.armigera* species and to describe the environment in which organisms exist. The prevailing weather conditions (minimum and maximum temperatures, morning and evening humidity) during *Rabi* (y season) 2002-03 were found to be similar to *Rabi* 1993-94 ($p< 0.01$). Therefore, Dymex was run for 1993-94 and 2002-03 *Rabi* season. Observed peaks of *Helicoverpa* incidence in 1993-94 coincided with the peaks simulated using Dymex. The validity of the model was verified by comparing the expected and observed moth populations during *Rabi* 2002-03 seasons. The expected number of adults were statistically non-significant ($P=<0.01$) with that of observed ones. Assuming that the weather conditions during the rest of 2003 might follow trends similar to those in 1994, incidence of *Helicoverpa* was predicted for the *Kharif* (wet season) 2003 season using weather file of 1994. The model was later validated using weather file of 2003 *Kharif* season. The peaks of *Helicoverpa* incidence again coincided with simulated ones ($p < 0.01$).

Humidity and host-plant as important factors influencing the pre-imaginal development of fruit flies (Diptera: Tephritidae)

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In Reunion Island, four species of fruit flies (Diptera: Tephritidae) cause damages on fruit crops namely the Mascarenes fruit fly, *Ceratitis catoirii* (Guérin Mèneville), the Mediterranean fruit fly, *C. capitata* (Wiedemann), the Natal fruit fly, *C. rosa* Karsch and the recent invasive peach fruit fly, *Bactrocera zonata* (Saunders). The influence of four important host fruit (guava, *Psidium guava* L., strawberry guava, *Psidium cattleyanum* Sabine mango, *Mangifera indica* L. and Indian almond, *Terminalia catappa* L.) on larval development and of a range of relative humidity on pupal development and the influence have been studied in the laboratory. Larval host had strong effects on survival, developmental times and pupal sizes (that possibly have consequences on fecundity of the female in the same generation). Humidity, especially low levels (30 and 50 %RH), had very important effect on survival depending on the fruit fly species. Indeed, *C. capitata* and *B. zonata* showed a high tolerance to dryness contrary to *C. catoirii* and *C. rosa*. These fruit fly species live in different ecological niches that can overlap. Results of this study are discussed according to the current distribution of these four fruit flies species in Reunion Island. Predictions can be also made on the future distribution of the invasive peach fruit fly.

Diversity, structure and composition of flying insects captured in fragment and continuos forests in the high Andean forest on the South West Sabana de Bogotá (Colombia).

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The structure and composition of flying insects in parcels of high Andean forest fragments and continuous was studied. A description of the morphotypes captured in omnidirectional and Malaisse traps was made. Species richness was measured (richness, estimators and Margaleff index) and Jaccard and Bray-Curtis similarity indexes were used to compare the composition by localities and traps; distribution models were used like structure descriptors. No differences were found between the insects collected in parcels of continuous forest (San Cayetano and La Selva) and those of fragmented forest (Miralejos and Silencio). The composition varied according to the used traps, thus being complementary methods, and the species richness differs between the four sites, being San Cayetano the site with the greater richness and La Selva the less. The richness estimators showed that the sampling was of low efficiency and logarithmic serial showed to be the most appropriated model. For Omnidirectional traps, Scolytidae (Coleoptera) was predominant, and for Malaisse Cecidomyiidae, Sciaridae and Phoridae (Diptera). The abundance of the community varied temporarily. One concludes that the communities partially reflect the differences attributable to the characteristics of the degree of conservation of forests and fragments.

Edge effect in the structure and composition of papilionoideos (Lepidoptera) of Andean forest at Sabana de Bogotá

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The composition and structure of the butterfly community to different distances from the edge was studied in two areas of high mountain forest. Four transects were drew toward forest respect to the edge (-60 to 150m), with one sampling station each 30 m; a control transect was located to 300 m in the forest interior. Traps, net and visual observation (at a perpendicular transect of 50 m by station) were used. 54 species were obtained (n=1346) in an efficient sampling (80% Chao 1). The edge showed the nearest value richness to the expected. The composition and abundance were organized in three groups of stations (external, edge and internal) by Jaccard and Bray-Curtis index. The richness and the diversity was greater in the edge according to observations and captures, the traps don't have differences in the gradient. The distribution of abundances followed the geometric, logarithmic and broken-stick models (external stations, edge and interior respectively). For butterflies, the edge effect positively penetrates until 60 m like minimum, affecting the richness and diversity of the edge; the forest act like a source of species for the edge. Useful species were identified to evaluate the penetration of the edge effect in Andean Forest.

Life table and demographic parameters of the "lettuce aphid" *Nasonovia ribisnigri* Mosley

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The objective of this study was to evaluate the development, survivorship and fecundity of *Nasonovia ribisnigri* (Mosley) at seven constant temperatures (8, 12, 16, 20, 24, 26, 28ºC) under laboratory conditions. The results obtained show that the proportion of alatae increased with increasing temperatures (T), remaining below 7% at T£16ºC and increasing to 40-57% at T³20ºC. The highest developmental period of apterae was obtained at 8ºC (31.5 days) and the lowest was at 26ºC (6.2 days), while the developmental time of alatae was approximately one day longer than for apterae at each of the constant temperatures used in the study. The survivorship of immature stages was always above 90% at the temperature range of 12-26ºC. However, the survivorship decreased dramatically at 28ºC (15%). The largest intrinsic rate of natural increase, r_m occurred at 24ºC (0.329) and the lowest at 8ºC (0.070). Unexpectedly, no nymphs were produced by adult morphs at 28ºC. The highest gross reproductive rate (åmx) and net replacement rate (*R*o) of *N. ribisnigri* occurred at 20ºC.The minimum and maximum developmental thresholds for apterae and alatae were calculated to develop a degree-day based model for this aphid species.

Population dynamics of aphid species colonising lettuce crops in Central Spain and identification of their associated parasitoids

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Aphid species colonising lettuce crops and their associated parasitoids were monitored during the spring and autumn growing seasons of 2001 and 2002 in Central Spain (Navalcarnero, Maid). Plants were sampled randomly following a zig-zag pattern by visual inspection at weekly intervals. The most abundant aphid species in both spring and autumn crops was *Nasonovia ribisnigri*, reaching the following peak densities at the end of each growing season: 80 individuals/plant in spring and 50 individuals/plant in autumn of 2001, and 132 individuals/plant in spring and 18 individuals/plant in autumn of 2002. The second major species present was *Macrosiphum euphorbiae*, reaching its peak density at the beginning of spring (2 and 0.3 individuals/plant in 2001 and 2002, respectively). In autumn, however, *Aulacorthum solani* was the second most abundant species reaching a peak density of 11 and 5 individuals/plant in 2001 and 2002, respectively. Three species of parasitic wasps (Hymenoptera: Aphidiinae) were identified from mummies of *N. ribisnigri* collected in lettuce crops: *Aphidius hieraciorum*, *Aphidius ervi* and *Aphidius colemani* . This is the first citation of *A. hieraciorum* acting as a parasitoid of *N. ribisigri* in Spain.

Ecological succession in bovine cattle dung pads - 60 years after Carl Mohr.

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The study of ecological succession in bovine dung pads is important for they are considered temporary microhabitats used by several insect species as ovipositing and feeding places. The objective of this research was to study the ecological succession occurring in dung pads from its excretion until they are not attractive to fimicolous insects anymore. Freshly excreted pads were placed in a pasture at 9:00 AM (F1), 9:00 PM (F2), 3:00 AM (F3) and 3:00 PM (F4), and insects were collected thereafter. More than 6,700 insects were collected and identified, mainly in Sepsidae, Sarcophagidae, Muscidae, Fanniidae and Chloropidae (Diptera), as well as Histeridae, Staphylinidae and Scarabaeidae (Coleoptera) plus Pteromalidae (Hymenoptera). The majority of the species visited the pads in higher numbers the first day after deposition. Paracoprids and endocoprids (Scarabaeidae) were mostly nocturnal; Sepsidae and Muscidae were most active in the morning and Sarcophagidae and Staphylinidae species in the afternoon. There were both diurnal and nocturnal Histeridae species. Muscidae and Staphylinidae were most abundant in F1 pads, paracoprids in F2 pads and Sepsidae in F3 pads; in endocoprids results varied. F3 pads took the longest (cerca 9 days) and F2 the shortest time (cerca 4 days) to be decomposed by Scarabaeidae.

Impact of the introduction of *Digitonthophagus gazella* (Diptera, Muscidae) on the native fauna of coprophagous Scarabaeidae in Brazil.

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Dung pads are a food source to many insects, mainly Coleoptera, Diptera and Isoptera, including several nematodes and flies associated with cattle, especially the horn fly, *Haematobia irritans*, one of the main pests of Brazilian cattle. Coprophagous Scarabaeidae (CS) while burrowing pads improve nutrient cycling, soil structure and forage growth for cattle, besides acting as efficient biological control agents of flies and nematodes of veterinary importance. *Digitonthophagus gazella* (DG) is an exotic CS to Brazil, and it was first registered at the UNESP Farm in Selvíria (Mato Grosso do Sul state) in 1993. Since 1989 CS are being weekly collected in a light trap in order to evaluate the impact of DG on the native fauna of CS. The diversity of species dropped sharply after DG invaded the area, which increased slightly three years later. For the majority of paracoprid species there was a reduction in the population density after DG was detected, while for some endocoprid species it increased. Some paracoprid species, considered common before DG colonized the area, are either not trapped anymore or in only small numbers. Ten years after DG invaded the Farm, it is still not clear if a new equilibrium was reached.

Dispersal kernels, matrix and marsh fritillary

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Two different dispersal kernels most frequently used in research on insect movements are the Negative Exponential Function (NEF) and the Inverse Power Function (IPF). In both, dispersion decreases exponentially with distance, but IPF has a wider tail, allowing for active long-distance dispersal. Using data from a metapopulation of the endangered *Euphyyas aurinia* butterfly in the Czech Republic, we tested how robust are the functions against reductions of data set, e.g., due to variation in sampling effort. Random removal of individuals, days, and both showed that slopes of the IPFs did not change when as much as 3/4 of data were deleted. IPF is thus superior for quick mobility assessments. We also asked how do matrix and quality of patches affect inter-patch movements in the metapopulation. In both sexes, movements were positively associated with a high number of individuals in original patch and absence of barriers (hedgerows), and negatively with the distance between patches. In addition, males were attracted to patches with many conspecific males, and females to patches rich on larval host plant.

Relationships of vegetation and soil dwelling Diptera colonizing initial succession stages

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This contribution summarizes the results of several research projects dealing with the initial stages of secondary succession in old fields, and areas disturbed by industrial use, post fire succession and driven succession used for restoration of species rich meadows in various part of central Europe. Soil dwelling Diptera are frequently abundant in these situations, and in many cases they dominate the soil macro-fauna. Some Chironomida (*Smittia atterima*) and Tipulidae (*Tipula scripta* and *T. paludosa*) and some other species are often more abundant in disturbed places than in the surrounding undisturbed landscape. Sites with a high abundance of chironomid larvae usually had little cover of higher plants, although areas with sparse vegetation were often preferred over completely bare sites. Tipulidae prefer sites with annual or weeds. Food resource analyses indicated that the presence of attractive food sources such as terrestrial algae, in Chironomidae and young seedling in Tipulidae are the main reason for preferential use of these habitats. Colonization of these optimal habitat have large impact on colonization sub-optimal habitat in the same site. As cover of optimal habitat increase density in sub-optimal habitat increased as well. On the other hand sub-optimal habitat may by in some situation more resistant to seasonal drought population in theses sub-optimal habitat may be important to overcome adverse environmental fluctuation.

Propagation-status of the fungal symbiont of a woodwasp, *Urocerus japonicus* (Siricidae) inoculated in living tree in the wood of Japanese cedar after felling

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Most woodwasps are symbiotically associated with *Amylostereum* fungi; female adults inoculate the fungi during their oviposition in host trees, and the larvae can digest wood with the aid of symbiosis. In earlier studies, we clarified females oviposit selectively in fresh felled trees that are presumed to be suitable for the fungus-propagation, and also clarified *U. japonicus* a fungus-carrying woodwasp can utilize the fungus which had already propagated in old felled trees. However, the inoculation-season in living trees suitable for the woodwasp-propagation has never been clarified. Thus, we conducted fungus-isolation to evaluate the symbiont-propagation status in inoculated trees. In July, we inoculated the fungus artificially in 6 living Japanese cedar trees (July-trees); 2 trees were felled in August and other 4 trees were done in November in the same year. Also in October, we inoculated the fungus in 2 living trees (October-trees), they were felled in November. In the next summer, fungi were isolated from all trees. In July-trees clear discoloration was recognized, while in October-trees little discoloration was done. In July-trees, the fungus propagated mainly within discoloration-areas, while in October-trees it propagated widely irrespective of discoloration-areas. These results indicate that trees inoculated in fall would be suitable for the woodwasp-propagation.

Sex ratio, stage distribution and larval interactions in *Chrysomya albiceps*

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Chrysomya albiceps is an exotic blowfly species, which differs from others in several aspects. It is a species with a naturally biased sex ratio and exhibits cannibalistic and predatory behavior over larval stage. In this study, we evaluated the impact of changes in sex ratio on the dynamic behavior of C. albiceps in a density-dependent mathematical model, analyzed the stage distribution by using the Leslie matrix model and investigated cannibalism and intraguild predation with functional response and interaction models. Bifurcation diagrams with periodicity windows between chaos regimes were found when sex ratio was evaluated. The stage distribution stooped changing after 40 generations, suggesting that the populations tend qualitatively towards stable stage distribution. No cannibalism occurred during the first and third larval instars of C. albiceps, but it was evident over the second instar. The functional response on second instar larvae tended to Holling type II. Investigating the intraguild predation, the flow of trajectories indicated the complete elimination of C. macellaria by C. albiceps and the coexistence between C. megacephala and C. albiceps.

Biology, dispersal and manner of damage by *Caryedon serratus* (Col.: Bruchidae) on *Prosopis cineraria* in Bushehr -Iran.

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Caryedon serratus is the most important pest on *Prosopis cineraria* because it causes the most damage in coastal areas from Kangan to Bandar-abbas. *C. serratus* has studied between (1997-1999) at Nature and Laboratory in Bushehr province. Significant damage was observed to seeds in June and November in nature. The damage was calculated at 20 to 30% on the seeds on the trees and 70 to 90% on the seeds under the trees on the ground. I observed oviposition to occur with mostly solitary eggs being laid and less frequently in a group (3-5 egg) on seeds and pod surfaces. After hatching, larva penetrated the seeds and destroyed them. Then, became pupa in the seeds and adults made exit holes on surface fruits. The eggs were elliptical and white by an 0.5 mm width and 1 mm length. the developed Larva were with 2 mm width and 6 mm length. Over wintering by larvae, pupae and adults was observed. over summering by adults on cover crop or under the sheath was observed. During the summer, these species were generated quickly and overlapping observed. *C. serratus* has 5 - 6 generations in a year. The life cycle was recorded 45 days at Laboratory (25úc-29úc) between June, July and August. This cycle was shorter in the Nature. *C. serratus* distribution was observed mostly at coastal lines in Bushehr province.

Potential indicators of biodiversity changes in a Mediterranean area subject to agriculture, forestry and livestock uses

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This work is part of the European Project "Biodiversity Assessment Tools" whose main objective is to assess changes in biodiversity. Butterflies, birds, carabids, springtails, soil macrofauna, plants and lichens were studied along a land use gradient. The study area was a typical Mediterranean with the natural vegetation comprising holm and cork oaks. The selection of the sites was made in the basis of information obtained on the area through remote sensing methods. Sampling was carried out in the Cabañeros National Park and its surroundings (Spain), during the years 2001 and 2002. For the land use gradient we selected six plots, with the natural forest in one side to the cultivated land on the other. The results are based on the analysis of 148000 specimens belonging to 1100 different taxa. The land use intensity affects the species richness and diversity in different ways in the different studied groups. The highest habitat diversity, typically found in areas with different land uses (agriculture, grazing, forestry) but without predominance of a particular one, favours the richness of birds, carabids and plants. Areas with natural forests show highest richness values of springtails, butterflies and lichens. Birds, plants and carabids would be adequate to assess the effect of agricultural or stock grazing uses. Butterflies, lichens and springtails are more relevant to assess the effect on biodiversity produced by forestry uses.

Genetic population structure of *Calathus erratus* (Carabidae) in postmining landscapes in Lower Lusatia (Germany)

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Colonization by arthropods of former coal mining areas of Lower Lusatia (Germany) was studied based on a comparative analysis of mining sites of different age and distance from undevastated areas including adjacent former military training areas. In the SUBICON project (www.subicon.de) investigations on the population structure of the carabid beetle *Calathus erratus* were carried out using AFLP method. We want to know whether the observed population differentiation was due to isolation by distance or different age of sampling sites. At the youngest sampling site (about 14 years after dumping), beetles were collected at different distances (1km, 5km) from the starting point. Further sampling sites comprise two older former coal mining sites (27 years, 5km and 56 years, 30km) as well as two former military training areas. First results with neutral molecular markers (AFLP) and AMOVA showed significant spatial genetic structure in that 98% of genetic variability resided within populations (only 2% among populations, global Fst=0.013). A Mantel test shows no correlation between genetic distances to geographic distances (Mantel test: p = 0.66). Colonization of devastated areas seems to take place rather quickly from surrounding biotopes. In fact we found at the youngest sampling sites 10% winged beetle individuals.

Characteristics of dipteran larvae infestations of marine turtle (*Caretta caretta, Chelonia mydas, Natator depressus*) nests and the impacts on reproductive success in central Queensland, Australia

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Dipteran larvae are known to infest marine turtle eggs, however, the few studies that have been conducted on this topic have presented conflicting conclusions as to the role of the larvae in the nest. This study will focus on the identification of the species of Diptera that infest green, flatback and loggerhead turtle nests in central Queensland, the role they play in the nest (detrimental, neutral or beneficial) and the subsequent consequences of that role on turtle hatch success. The three mutually exclusive roles listed above will have markedly different effects on the subsequent hatch success of affected marine turtle nests in central Queensland. If the larvae function in a detrimental role as predators, they will negatively affect hatch success. A neutral role would imply that they simply devour dead material accumulating in the nest. Lastly, the larvae could be functioning in a beneficial manner by removing decomposing material that might otherwise lead to fungal or bacterial infection within the nest, and thus they may positively affect hatch success. This study will provide useful information for the management of marine turtles in the central Queensland region.

Composition and structure of bark and wood-boring pest communities in Belgian beech (*Fagus sylvatica*) stands, in relation to dead wood stock.

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Because of economic reasons and of forest hygiene measures, dead wood and overmature trees (DWOT) are almost systematically removed from woodlands in Wallonia (Southern Belgium). Consequently, the average amount of dead wood in Walloon deciduous forests is less than 5 m^3/ha, whereas it would be of several tens of m^3 per ha if those forests were left unmanaged. There is nowadays growing awareness of the crucial roles that DWOT play in forest ecosystems and of the need to restore those habitats. Nevertheless, because of misunderstanding and lack of adequate information, they are often considered as pest sources and threats to timber production. Since complex relationships link xylophagous pests, their antagonists, saproxylic communities and DWOT, leaving a part of the latter in deciduous forests would however not necessarily imply a significant increase of xylophagous pest population levels. In this context and as part of a more global study, we present and discuss the composition and structure of xylophagous pest communities caught in 2002 in five beech (*Fagus sylvatica*) stand pairs (fauna surveyed with eight unbaited window-traps per stand). Each pair consists of a stand with few DWOT and another stand with similar characteristics but with high quantity of DWOT.

Patterns of population fluctuations of the mulberry tiger moth, *Lemyra Imparilis* (Butler): effect of predation by the specialist carabid predator, *Parena perforata* (Bates)

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Population changes of the mulberry tiger moth, *Lemyra imparilis* (Butler) (Lepidoptera: Arctiidae) and its carabid predator, *Parena perforata* (Bates) (Coleoptera: Carabidae) were surveyed at two disjunct study sites on Kii Peninsula, western Japan, from 1989 to 2003. The population fluctuation of *L. imparilis* showed a pattern in which the density approaches peaks slowly but collapses rapidly. The analysis of population growth rates showed that delayed density-dependent factors act as important roles regulating *L. imparilis* populations. At one study site, the number of *P. perforata* per prey colony changed closely with the density of *L. imparilis*. During the decline phase of *L. imparilis*, *P. perforata* was observed in over 80% of the prey colonies and the predation effect acted strongly. At another site, however, the carabid beetles were very few. At this site, the peak shape of *L. imparilis* population change was flatter than that of another site having sharp and angular peak. In addition, the amplitude was ca. one-third that at another site. It was suggested that the predator-prey interaction constitutes an important factor in explaining the different behaviors of *L. imparilis* populations at different localities.

Spatio-temporal dynamics of dung beetle communities in pastures

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Dung beetles (Coleoptera: Scarabaeidae) play an important role to decompose cattle droppings in pastures. The rate of the decomposition should depend on the dynamics of dung beetle community in the pastures. We surveyed dung beetles by a dug-bated trap in 17 pastures located in the basin of River Naka in Tochigi prefecture, Japan, once a month from May to October for three years to elucidate the dynamics of dung beetle communities. Twenty-five species were collected (21 were used to analyze). The community varied among pastures and years in terms of species richness, abundance and structure. We analyzed the data by principal component analysis (PCA) using a software ADE-4 (Thioulouse et al., 1997) to evaluate temporal and spatial dynamics of the communities. Ordinal PCA indicated that regional abundance and yearly change in abundance of the beetles mostly explained the variability of the communities. The spider graph representing variability of factor scores among pastures and years on Z1 - Z2 plane predicted that the former was larger than the latter. Further analyses by within- and between- groups PCA demonstrated that the spatial dynamics was much more important than the temporal one. Environmental and biological factors seemingly influenced the spatio-temporal dynamics of the communities.

Parasitoid's switching predation enhances persistence in two-host one-parasitoid experimental system

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Effect of predation on the persistence of competing prey species were examined by laboratory experiments. Two bruchid beetles, *Callosobrucus maculatus* and *C. chinensis* were used as the competing prey species, and the parasitoid of these beetles, *Anisopteromalus calanae* was introduced as its common predator. The currently existing theoretical models suggest that persistence of systems depends on foraging strategy of the predator; random or switching predation (Murdoch and Oaten 1975; Post et al. 2000). In this study, we aim to determine how the persistence and dynamics of this system are affected by predation. The time until extinction of either of prey species was observed by the long-term multi-generation experiment, and the foraging behaviour of *A. calanae* was also examined by short-term experiments. In all long-term experimental replications without wasp, *C.chinensis* was competitively excluded. On the other hand, significantly longer persistent time was observed in the presence of wasps, in which alternate population oscillations were observed in two prey species. The short-term experiments suggested that *A. calanae* learned hosts which they had oviposited and preferred it more when parasitising. This behaviour results in positive frequency-dependent predation, and causes persistence and the dynamics pattern in this system with parasitoid.

The significance and variability of predation of weed seeds in an agricultural landscape in Western Australia.

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Understanding the fate of weed seeds over summer in agricultural fields is not well understood, but is necessary in order to develop possible alternatives to weed control – particularly when considering the increasing failure rate to herbicides. It is widely accepted that granivory plays a role in regulating plant populations and is a major contributor to seed loss in natural ecosystems. Despite this, the relationship has not been thoroughly researched or proven to significantly reduce seed banks of weed species in agricultural environments. Three years of study in the Western Australian Wheatbelt has shown that: (1) predation levels are higher for annual ryegrass than for wild radish and can vary from 0 to 100% within the same field; (2) predation levels tend to be higher on the edges of field rather than in the centre, but only when using artificial seed populations; (3) when using natural weed populations, a threshold in predation level occurs; (4) ants play the dominant role in seed harvesting and were found to consume seeds, rather than disperse them; (5) the presence of particular ant species can possibly be used as a predictor of weed seed predation levels; and (6) crop residue types and tillage practices can affect the level of seed predation.

Bottom-up cascade in a tri-trophic system: different impacts of host-plant regeneration by cutting on performance of a leaf beetle and its natural enemy

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The present study examined the effects of artificial cutting of a willow on performance of the willow leaf beetle and predatory ladybird through trophic levels. Laboratory experiments showed that performance (survival rate, developmental time, and adult mass) of the willow leaf beetle, *Plagiodera versicolora* Laicharting, was improved by feeding on leaves of cut willows, comparing to leaves of uncut willows. Performance (developmental time and adult mass) of a predatory ladybird, *Aiolocaria hexaspilota* Hope, was also improved by feeding on the leaf beetle larvae that fed on leaves of cut willows, compared to that fed on leaves of uncut willows. In comparison of improved performance parameters between the leaf beetle and ladybird, regenerated willows more shortened developmental time of the willow leaf beetle than that of ladybird. This indicates that impacts of willow cutting on insect performance differed between second and third trophic levels.

Nitrate and soluble carbohydrates stimulate mosquito production from container habitats of the tree hole mosquito, *Ochlerotatus triseriatus*.

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Most habitats of container breeding mosquitoes are dependent on allocthonous input of nutrients. One mechanism of nutrient input into tree hole ecosystems is via stemflow during storm events. Stemflow contains soluble organic and inorganic nutrients and also may serve to flush out potentially inhibitory metabolites or re-suspend particulate nutrients from sediments. We investigated the effects of flushing and soluble nutrient additions on *Ochlerotatus triseriatus* development in microcosms using a factorial experimental design. Both simulated stemflow flushing and nutrients strongly affected mosquito production, however, there was some evidence of interaction between the two factors. In general, the additions of nitrogen (as nitrate) or soluble carbohydrate (as glucose) had positive effects on mosquito production, but flushing had negative effects. Additions of phosphorous (as phosphate) had no effect. Interestingly, the addition of nitrogen enhanced leaf mass loss and contributed to higher soluble carbohydrate levels in the water column, most likely via stimulation of the fungal community associated with leaf detritus. The results are consistent with an indirect effect of nutrient inputs on mosquito larvae that is first channeled through microbial community components.

Ants of the *Formica rufa* group affect nutrient dynamics and forest growth in boreal forests

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Mound-building red wood ants (*Formica rufa* group) are proposed to be a key-species in boreal and subarctic forests of Fennoscandia. The aim of our study is to understand the role of wood ants in the functioning of boreal forest ecosystems. Large-scale disturbances, like clear-cutting, can make wood ants abandon their mounds. Our study areas are located in eastern Finland in medium-rich coniferous forests of four age classes after clear-cutting. Changes in mound number, size and spatial organisation, carbon (C) and nutrients pools will be quantified in each ecosystem. We are assessing annual C and nutrients ant mound inputs (litter, honeydew, invertebrates) and outputs (CO_2, nutrient mineralization). Based on preliminary calculations, the ant-aphid symbiosis can significantly reduce the growth of coniferous trees. The impact of the ant-aphid symbiosis on the growth of conifers is also being measured. Preliminary results indicate that ant mounds are bigger and contain more C and nutrients in older forest stands than in younger stands. The C and nutrient contents of ant mounds are small compared to other pools in forest soils but locally the accumulation and fluxes of C and nutrients in ant mounds can be important to ecosystem processes.

Genetic structuring of western corn rootworm populations in the US based on microsatellite loci analysis

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The western corn rootworm (*Diabrotica virgifera* LeConte) (Coleoptera: Chrysomelidae) is a major corn insect pest in the United States. Understanding gene flow and dispersal patterns of this insect is essential to designing efficient strategies of monitoring for resistance to *Bt* toxin in genetically modified corn. Microsatellite markers for population genetics studies of the western corn rootworm have been developed using an enrichment method. As of this writing, 24 microsatellites have been prescreened in automated genetic analysis system with fluorescent-labeled PCR primers. Of these, nine di-nucleotide and two tri-nucleotide repeat microsatellites have been confirmed as polymorphic. These polymorphic microsatellite loci are being surveyed to obtain genetic variation among western corn rootworm populations of 540 individuals sampled from 9 locations across eight U.S. states. The results will be used to infer the magnitude and pattern of genetic differentiation among western corn rootworm populations on a broad geographic scale, as well as genetic structuring of this insect.

The assessment of toxicity of 4-nonylphenols on the *Chironomus flaviplumus* using molecular biomarker

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The assessment of the contamination level consists of comparing the biochemical substances in the macroinvertebrates. Many chemical materials induce an adverse effects on aquatic ecosystems. Alkylphenols selected among the chemical materials in this investigation, because it detected at the study sites. Among the alkylphenols, bisphenol-A and 4-nonylphenols were detected in these samples. Environmental samples were analysed by GC-MS method. Also biochemical marker investigate by the 2-D electrophoresis and MALDI-TOF mass spectrometric analysis. Our studies showed that up-streams as well as downstreams in the study sites have been contaminated by 4-nonylphenols. The distribution of tolerable species to pollution such as Diptera, were in accordance with the higher concentrations of alkylphenols. Fourth-instar *Chironomus flaviplumus* larvae were exposed to the 4-nonylphenols (0, 1, 10, 100 /L) for 24h at 25°C. After acute toxicity test, treated samples showed ascension of vWF activity to the larvae *Chironomus flaviplumus*. The vWF is related to blood coagulation. This result suggesting that vWF is possible for use as a biomarker of toxicant exposure or effect in Chironomidae.

Effects of climatic change on the spruce bark beetle *Ips typographus*, an economically important forest pest

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The spruce bark beetle is the most serious pest of mature spruce trees in Eurasia. At low population densities it breeds in weakened or newly dead trees, but at high densities it becomes a tree killer. The exact factors triggering outbreaks are not fully understood, but climatic variables are important candidates. Populations in SE Norway have been monitored since 1979, and using these data we want to (1) explore the role of climate in outbreaks of the spruce bark beetle in space and time, (2) estimate the distribution of bivoltism under different scenarios of climate change, (3) estimate the risk of forest damage under different scenarios of climate change, and (4) describe forest management practices that will reduce the impact of beetle outbreaks triggered by climate change. Large windfall events appear to be a major instigator and synchronizer of beetle outbreaks in areas subjected to regionalized weather systems. A phenological model sets beetle development as a function of local temperature. This model will be compared to local reports of bivoltism, and finally used for predictions of bivoltism under various scenarios of temperature change.

Dispersal of West Indian sweet potato weevil, *Euscepes postfasciatus* (Fairmaire), in sweet potato field

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West Indian sweet potato weevil, *Euscepes postfasciatus* (Fairmaire), is one of the most important pests of the sweet potato, *Ipomoea batatas* L., in tropical and subtropical areas including the southwestern islands of Japan. In this study, we made two 5 by 5m square sweet potato fields scaffolding for observation not to disturb the dispersion of this weevil. Forty-eight sweet potato tuber traps were set inside and around each field. We released about 2,000 mated and unmated male weevils into the center of one field. Similarly about 2,000 mated and unmated female weevils were released into the center of another field. These weevils were sterilized before releasing. We marked each weevil with oil-based marker to be able to distinguish mated and unmated, male and female respectively. We checked the traps everyday for 1 week and 15th day. On 15th day, we collected weevils and checked the seminal receptacle of females to confirm the mating. As a result of this experiment, we found that males and mated females dispersed in the field uniformly and moreover went out from the fields, on the other hand, unmated females distributed center of the field contrastively.

Effect of aerial pesticide application on ant foraging activity

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Aerial pesticide application is main control method for pine wilt disease in Korea. Fenitrothion, used for the application, is the chorinesterase inhibitor. This suggests that the aerial pesticide application invoke retardation in activity as well as death of arthropods. This study is aimed to find changes in foraging behavior of ants due to aerial pesticide application. Foraging activity of ants had been observed by using baiting cards across the period of the three aerial pesticide applications in June. The activity was significantly decreased after the pesticide application. Such behavior retardations were different among ant species. As days passed, it recovered gradually and became normal state in 16th day after the application. Feeding behavior of ant species were affected by the presence of other ant species. *Paratrechina flavipes* inhibit *Componotus japonica* in feeding food, and the reverse was true. Interestingly*, Monomorium chinense* was stimulated in feeding by the presence of *Paratrechina flavipes*.

Model application to egg development of aquatic insects – 1. ***Ephemera*** **spp.**

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Because the water system was more stable than terrestrial system in the temperature aspect, temperature related development models would be more useful to estimate the rate of development in aquatic insects than in terrestrial insects. 5 descriptive models on Arthropods, 3 Logan's, Stinner's and empirical model according to the Arrhenius' rule, were examined to evaluate their applicability to aquatic insects. In this study models were applied to egg development of two *Ephemera* spp., because no other factors, except temperature, could be thought to have effects on egg development. Among models, Logan's original model showed best description on the egg development while Stinner's was the worst. As known, Stinner's model did not explain the abrupt decrease of developmental rate after the optimal temperature although this showed good fit to ascending part. Logan's sigmoid model showed two problems. One is the forced sigmoid pattern in ascending developmental rate, which caused large variance. Another problem is the overestimation of maximum developmental rate. The overestimation was also shown in application of empirical model. Though empirical model showed relatively good-fit, this had too many parameters. Logan's modified model did not show good-fit compared with two other Logan's, but had advantage in estimating the base temperature.

Temperature adaptation in Arctic Collembola: thermal reaction norms of egg development rates.

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Thermal reaction norms, expressed as temperature dependent development rates of egg development, have been studied in a number of high arctic collembolan populations. Four related species co-existing in the same study site on Svalbard were compared. In all three species the development rate increased up to 25°C (from 5°C as the lowest temperature). However, the species differed in the slope and shape of their thermal response curves. In the lower temperature range (5-10°C) they were virtually overlapping, but at higher temperatures the increase in rate was much slower in two typical high arctic species than in the two other with a much wider distribution in both temperate and cold areas. Thus, although a warmer climate may have a positive direct effect on all species, the two high arctic ones appear to it relatively less and thus may get an indirect competitive disadvantage. Three species with wide distribution across climatic zones differed fundamentally in intra-specific variation of the thermal reaction norm along climate gradients (latitudinal ranges of ca. 20°), i.e. both increasing and decreasing in populations from increasingly colder environments. This is explained by differential time constraints in the northern populations depending on their phonological strategy and reproduction flexibility.

Effects of temperature on the development of *Aulacaspis murrayae* Takahashi (Hemiptera: Diaspididae)

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The effect of temperature on the development was determined for three stages of an armoured scale insect, *Aulacaspis murrayae* Takahashi (Hemiptera: Diaspididae), a key pest of orange jessamine (*Murraya paniculata* (Linn.) Jack) in Taiwan. The scales were reared on orange jessamine at 4 different temperatures (15, 20, 25, and 30°C). The results showed that an increase in temperature within limits accelerated the rate of development of all stages. The developmental duration of the scales from ovum to adult at 15, 20, 25, and 30°C were 78.2, 60.3, 49.31, 42.81 days. Temperature also influences the reproductive capacity of the scales. Lower temperatures increased egg production of the scales. A linear regression of developmental rate against temperature showed that there was a high correlation between temperature and growth rate for all stages ($r^2 = 0.8926$). Their low developmental threshold temperature (Tb) and thermal summation (K) were: 5.6°C and 174.6 DD in the egg stage; 6.8°C and 128 DD for crawlers, and 2.9°C and 284.3 DD for secondary instars, respectively.

Community dynamics of ground beetles (Coleopetera: Carabidae) in a successional forestry landscape

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Western European Lowland Heathland is a threatened habitat recognised by the European Community and the UK Biodiversity Action Plan. Much of this valuable habitat, especially for invertebrates, has been replaced by afforestation. Within the UK, distributions of a number of ground beetle species are restricted to Breckland, and the creation of Thetford Forest has contributed to losses of heathland in this area. A two-year survey has shown that clear-felled areas and ride margins within the forest have considerable value for heathland ground beetles. The ground beetle compositions through successional stages, age 0 to 5 years, are studied and also compared to that of mature forest (age 52 to 73 years). The effect of succession on ground beetles was then investigated at species level in relation to their ecology such as habitat preference and dispersal ability. The implications for conservation and forestry management of understanding the ground beetle dynamics in this landscape scale are also considered.

Infestation of *Adalia bipunctata* by a parasitic mite *Coccipolipus hippodamiae* (Acarina: Podapolipidae) in Moscow

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The mites of the genus *Coccipolipus* are ectoparasites of coccinellid beetles. The major *C.hippodamiae* host is two-spotted ladybird *A.bipunctata*. The adult beetles were collected in various places of Moscow, Russia. The seasonal dynamics of *C.hippodamiae* abundance was evaluated from May to September 2000 and 2002 by collecting and examining beetles weekly; the mites were counted on 20-50 *A.bipunctata*. A total of about 800 beetles were collected during the study period. Mite infestation of beetles was 14-25% in the beginning of May. The number of mites (larvae, males, females) was 1,7-8,1. In summer (June to August) mite infestation of *A.bipunctata* was higher than 50% (96,3-98,9% in 2000 and 50-90% in 2002), with average abundance of *C.hippodamiae* on one infested beetle about 30. The mites formed large stocks on some beetles. The maximum number of adult females collected from one beetle was 24, whereas the highest number of eggs – 690. There was no *A.bipunctata* sex preference in mite infestation. In September the occurrence and abundance of *C.hippodamiae* were significantly lower. The percentage of infested beetles fluctuated from 4 to 13 in different places in Moscow in 2000, 2002. The average number of mites on one infested *A.bipunctata* was 8,3-13,1. *C.hippodamiae* larvae dominated in summer, females – in September.

Summer raids of *Arocatus melanocephalus* (Heteroptera: Lygaeidae) in houses of Modena city (Italy): is climate change to blame?

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Arocatus melanocephalus is a small lygaeid performing its life cycle on elms (*Ulmus spp.*, Ulmaceae). Its presence has been recorded in many continental European regions. Its occurrence has been ascertained in Northern Italy since the last century; however it never elicited special attention until summer 2000, when it suddenly started to become a problem for the citizens of Modena (Emilia Romagna). Since that time, every summer on several occasions, thousands of adults of *A. melanocephalus* leave the trees to enter the houses, gathering in hundreds all over doors, windows, balconies, and penetrating inside the rooms, causing trouble and anger in the people. Among all the possible reasons for this phenomenon, we suggest that climatic changes observed in Modena during the last 4 years may have affected the behaviour and/or the population dynamics of this bug. In agreement with global warming signals, meteorological recordings for this city show a temperature increase of about + 0.8°C/100 year, and anomalous peaks were observed especially during spring months, likely in coincidence with crucial events in the life cycle of the insect. Studies on the reactions of different stages of *A. melanocephalus* to cold/hot temperatures are currently performed to provide further support to our hypothesis.

Prey suitability of *Myzus lythri* for generalist predators

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Understanding the interactions among the trophic levels that structure the purple loosestrife, *Lythrum salicaria* complex in invaded wetlands will contribute to management options and a better knowledge of invasive species. In 1992, *M. lythri* was observed feeding in abundant numbers on *L. salicaria* in Indiana in the North Central United States. The suitability of *M. lythri* nymphs and adults, for development and survival of *Harmonia axyridis* Pallas, *Coleomegilla maculata* (DeGeer) (Coleoptera: Coccinellidae), and *Chrysoperla carnea* (Stephens) (Neuroptera: Chrysopidae) was quantified by measuring preimaginal development and survival. Individual first instars were assigned to one of two diets: *Acyrthosiphon pisum* (Harris) (Homoptera: Aphididae) or a diet of *M. lythri*. The pea aphids, *A. pisum* serve as the control diet because other studies use these aphids as diets for generalist predators and the predators' development seems optimal. *Coleomegilla maculata* and *C. carnea* development was delayed and lower survival rate with *M. lythri* diet compared to *A. pisum* diet. *Harmonia axyridis* development was delayed with *M. lythri* diet but survival was similar to *H. axyridis* fed *A. pisum*. These studies may provide an insight of what is occurring in the wetland areas where these aphids and predators are present. Trophic interactions such as these may be affecting the density and biomass of purple loosestrife in the wetlands.

Habitat selection by the antlion *Myrmeleon bore* living in a sandy riverbed (Neuroptera: Myrmeleontidae)

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The pit-making antlion larvae *Myrmeleon bore* live in open sand in riverbeds. The spatial distribution pattern of their pits showed an aggregative distribution. We analyzed which environmental factors affect dominantly their spatial distribution out of a biotic factor (prey availability) and abiotic factors (sand temperature and particle size of sand). There was a significant positive correlation between the pit density and the ratio of medium sand to total weight of sand, while there was no significant correlation between the pit density and the temperature of sand or prey availability. Experiments on the oviposition behavior revealed that females of *M. bore* laid more eggs in fine or medium particle size sand than in coarse sand. This suggests that habitat selection by adult females is a major factor that causes the aggregation distribution of the pits, because the larvae rarely relocate their pits.

Shifts in habitat associations of dung beetles: climate change implications

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We examined whether dung beetle species shift their habitat associations (vegetation types) with different climatic conditions, using altitudinal and seasonal gradients as analogues of future climate change scenarios. Dung beetle assemblages in Picos de Europa (northern Spain) were monitored by dung-baited pitfall trapping in three vegetation types (woodland, heathland and pasture), three altitudinal zones (520-600 m, 950-963 m and 1230-1275 m) and during two different seasons (summer and autumn) in 1994. Most species shifted their habitat use on the altitudinal and seasonal gradient in response to changes in microclimatic conditions (temperature). They were generally associated with heathland and woodland (shaded habitats) at lower altitudes, but they were increasingly associated with pastures (open habitat) as altitude increased. There was also a greater association with pastures in the coldest season (autumn) and with heathland and woodland in the warmest season (summer). These results suggest that climate change could be a major factor affecting habitat selection in dung beetles. The research presented here supports the feasibility of using microclimatic gradients within a region to study the potential effects of future climate change on both species and communities.

Leaf selection of leaf-mining *Phyllonorycter* **moths in a deciduous oak** *Quercus dentata*

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Phyllonorycter leucocorona and *P. persimilis* are dominant leaf mining moths on a deciduous oak, *Quercus dentata*, in Sapporo, northern Japan. These moths overwinter as pupae and pass two generations in a year: females of the first generation oviposit in June after most ectophagous caterpillars have completed their development, and females of the second generation oviposit in August. Their larvae have two functional stages, the sap-feeding and tissue feeding stages. Here we report leaf selection of these moths. In this study, these two species were treated together, since they could not be discriminated at the sap-feeding stage. The density of their mines was negatively correlated with leaf size, but positively correlated with the degree of leaf damage by ectophagous insects. Survival of sap-feeding larvae was slightly higher in small and damaged leaves than in larger or less-damaged leaves, suggesting a weak preference-performance linkage. On the other hand, the analysis of distribution pattern of mines suggests that adult female moths do not prefer or avoid leaves that have been previously oviposited by congeneric females. In addition, survival of sap-feeding larvae was not affected by the mine density.

The rediscovery of a life cycle form, which has oviparae produced by apterous viviparae, and crossing trials between holocyclic clones in *Aphis gossypii* Glover (Homoptera: Aphididae)

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The life cycle form of *Aphis gossypii*, that apterous viviparae produced oviparae, has been discovered by Thomas (1968). We found the same form on *Fragaria grandiflore, Chrysanthmum morifolium* and *Lillium* spp etc. 7 different host plants in Japan. We investigated the inheritance of life cycle forms using GP1 clone from *Pyrus serotina*, the same life cycle form, and GHS clone from *Hibiscus syriacus*, that oviparae were produced by only alate viviparae (heteroecious form). We carried out crossing experiments by selfings and reciprocal crossings of GP1 and GHS. All of 27 F_1 clones in selfings of GHS were heteroecious form. 20 F_1 clones in selfings of GP1 were separated into 10 GP1 forms, one heteroecious form, and 9 anholocycles. 78 F_1 clones in crossing of GP1 (female) * GHS (male) were separated into 40 GP1 forms and 38 heteroecious forms. 90 F_1 clones in crossing of GHS (female) * GP1 (male) were separated into 53 GP1 forms, 35 heteroecious forms and 2 anholocycles.

Mortality factors affecting populations of sweetpotato whitefly, *Bemisia tabaci*, in a multi-crop system

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The sweetpotato whitefly, *Bemisia tabaci* is a major pest of world agriculture, causing damage by direct feeding, vectoring plant viruses, and excretion of honeydew. *B. tabaci* is a multivoltine insect with no quiescent stage and reproduces by moving from one host to another over the year. Thus, management on any one crop is inexorably tied to other crops and wild hosts surrounding it in space and in time. To better understand the mechanisms governing population development of this insect, whitefly "ecosystems" were established in three geographically and climatically distinct areas in Arizona. These systems consisted of a sequence of six representative hosts including winter broccoli, spring and fall cantaloupes, summer cotton, perennial alfalfa, various annual weeds, and the perennial ornamental, *Lantana*. Source and rates of mortality were quantified, *in situ*, on each host plant by constructing life tables. Factors causing mortality were the same in all hosts; however the contribution of each mortality factor varied among hosts and time of the year. Predation and dislodgement from the plant surface were consistently the largest sources of mortality. An understanding of the year round dynamics of this pest will greatly aid the development of better pest management strategies in all affected crops.

Host-tree decline and pulse eruptive outbreaks of *Creiis lituratus* (Hemiptera: Psyllidae) in *Eucalyptus dunnii* plantations

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Localised stressful conditions such as drought or water-logging have been shown to retard the growth of *Eucalyptus dunnii* planted in northeast New South Wales. During a two year field study in *E. dunnii* plantations we observed increasing densities of the leaf damaging psyllid, *Creiis lituratis,* over the autumn, winter and spring months. In most of the study plots the heat-related mortality of approaching summer led to a collapse of the *C. lituratis* population. When the capacity of host trees declined prior to the onset of high temperatures, density-dependent dispersal of the psyllid took place. Areas immediately surrounding hot spots experienced an influx of *C. lituratis* adults, with population development following pulse eruptive dynamics, and resulting in larger areas of severely damaged trees.

The biology of *Lissoderes pusillus* Hespenheide (Coleoptera: Curculionidae: Conoderinae) in *Cecropia* (Cecropiaceae) saplings

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The eggs, larvae, and pupae of *Lissoderes pusillus* Hespenheide occur inside the hollow stems of *Cecropia* saplings, but are usually only present in internodes not occupied by live *Azteca* ants. Eggs are deposited on the inner surface of the internode. Larvae feed on the parenchyma, the same food resource that is utilized by *Azteca* queens. Weevil larvae feed and move about in an upside-down position. Fleshy folds on the dorsum of the body are apparently utilized in locomotion. Larvae are usually restricted to just one internode, but are more likely to move to an adjacent internode in *Cecropia* species possessing a thin parenchyma layer. Pupae hang freely from the inner vertical walls of the internode. Two hymenopteran parasitoids were reared from larvae and prepupae: *Neocatolaccus* sp. (Pteromalidae) and *Heterospilus* sp. (Braconidae). *Menozziola* sp. (Diptera: Phoridae) was observed parasitizing *Azteca* queens. The high mortality of colonizing *Azteca* queens does not appear to be caused by insect parasitoids or by competition with *L. pusillus*.

Thaumastocoris australicus, **the bug turning green Sydney red.**

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Over the past six years a thaumastocorid bug, *Thaumastocoris australicus*, has had a devastating impact on some *Eucalyptus* species in the Sydney metropolitan area. The symptom of this infestation is now so widespread it is commonly called "winter bronzing". Initially this winter bronzing was found on two eucalypt species, *Eucalyptus scoparia* and *E. nicholii*, both very common street trees in Sydney. Infested trees display a reddening of leaves. Severely infested trees can die but usually are removed. Over the past two seasons another nine eucalypts have been verified as infested. Given the magnitude of the problem with respect to the bug's numbers and expanding host range, combined with the logistics of control, a study of the bug is being undertaken. A standard stratified technique was used to sample thaumastocorids at each aspect of three *E. nicholii* trees over 18 months and a comprehensive range of data has been recorded. These data include: sex ratios of adults; numbers and instars of nymphs; numbers and location of eggs; presence of parasitoids. Laboratory work has been conducted to elucidate aspects of the bug's life cycle.

Evolution of the mate recognition key stimuli (ultraviolet wing color) in the cabbage butterfly, *Pieris rapae crucivora*

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The female of the Japanese subspecies of cabbage butterfly, Pieris rapae crucivora, has been shown to have the ultraviolet (UV) color on the wings which is an essential component of the key stimuli for the mate recognition by the conspecific male. There occurs, however, no UV color in the female wings of British subspecies, P. r. rapae. This raises some interesting subjects to be tackled. One is where in the Eurasian Continent between the two countries the female wings with UV color evolved. The results of UV photographs taken through UV filter transparent only to UV light (300-400 nm) revealed that the female specimen from all the regions of the Continent but East China had no UV color, suggesting that female UV wing color should have evolved in East China. The other is whether there occurs behavioral isolation between the two subspecies due to the difference in the key stimuli. The results of the behavioral experiments showed that the males preferred each their own subspecies of females. The British subspecies of males, however, were strongly attracted by the short-day form of Japanese females with intermediately strong UV color. The behavioral isolation is, therefore, partial.

The effect of male killing on spatial and temporal distribution of the African monarch butterfly (*Danaus chrysippus*) population in Kenya.

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Danaus chrysippus (L.) (Lepidoptera: Danaidae) butterfly is the commonest, most polymorphic of all butterflies found in the old world tropics. The danaid butterflies are the most unique of all tropical species since they are known to use chemicals for protection against predators, thereby becoming prime models in mimicry complexes. A maternally inherited male killing bacterium (*Spiroplasma)* that infects the butterfly and kills their male during the early developmental stages and result in a female biased brood has been found in Kenya. Ordinary butterfly nets were used to collect the butterflies for analysis. The results show a steady decline in the number of males in the wild. The five sites sampled in Kenya show low hatch rates (53.13 - 86.95%). Seasonal results show that females dominate (71%) with *dorripus* form (67.9%) being the most common followed by *chrysippus* form (22.6%). Female butterfly population seem to outweigh their male counterparts although eggs are laid singly which suggests that the ecological conditions favouring male killers are quite widespread. From the results, the male population face significant depression. More research needs to be done on the spatial - temporal distribution and dynamics of male – killing *Spiroplasma* bacterium to model male *D. chrysippus* population trends.

Tyrophagus putriscentiae (Schrank) (Acaridae: Astigmata): a predator of *Lasioderma serricorne* (F.) (Coleoptera : Anobiidae) reported for the first time in tobacco stores.

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Studying the biological complex of *Lasioderma serriccorne* inside tobacco stores, during the years 2002 and 2003, the presence of the cosmopolitan mite species *Tyrophagus putriscentiae* preying on live *L. serriccorne* larvae was noted. During the above two-year period, samples of dried tobacco leaves infested by *L. serriccorne* were taken and the rate of parasitism by the mite was measured. The rate of *L. serriccorne parasitized* by *T. putriscentiae* depended on time and tobacco store, was about 20%. *T. putriscentiae* is reported for the first time as a predator of *L. serricorne* in tobacco stores in Greece.

Tephritoidea species infesting Myrtaceae fruits in the state of São Paulo, Brazil

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A field survey was conducted from August 1997 to January 2003 to identify the Tephritoidea diversity from 15 Myrtaceae species (including guavas) collected in the State of São Paulo, Brazil. A total of 38,386 fruits (203,93 kg) were collected from unsprayed trees. From 188 samples of fruits 25,162 puparia and 15,439 adults of Tephritoidea were recovered. *Anastrepha* spp. and *Ceratitis capitata* adults were obtained from 78,7% and 16.5% of the samples, respectively. About 95.8% of all adults collected were Tephritidae and 4.2% were Lonchaeidae. Only 1.7% of Tephritidae adults corresponded to *C. capitata*. From 14,540 specimens of *Anastrepha* we obtained 6,642 *Anastrepha* females identified as: *A. fraterculus* (87.7%), *A. bistrigata* (5.7%), *A. obliqua* (3.7%), *A. sororcula* (2.4%), *A. zenildae* (0.2%), *A. striata* (0.2%) and *A. turpiniae* (0.1%). All seven *Anastrepha* species were recovered from guavas. Mean infestation indices were 0.66 puparia of Tephritoidea/fruit and 123.4 puparia/kg of fruits. The samples infested by Tephritoidea was higher than 88%.

Spatial distribution and dispersal of *Heteronyx crinitus* and *H. dimidiata* in Tasmania.

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High populations of *Heteronyx dimidiata and H. crinitus* can cause serious defoliation to young *Eucalyptus nitens* in Tasmania. An understanding of the distribution and dispersal in the soil of these species is important for determining critical thresholds and population dynamics. This study investigated the homogeneity of their distribution around the host's root-zone. Destructive sampling, which involves removal of entire tree as a monolith can link residual population to a host. Soil populations were investigated by destructive sampling 120 random trees over 12 rows. *H. dimidiata and H. crinitus* populations coexisted, but were present in different densities. Site characteristics, including tree size, leaf totals, general condition and winow proximity were found to be of low importance. A species ratio of 1:8.4 (*H. crinitus*: *H. dimidiata*) was found. It was concluded that *Heteronyx spp*. in an over-wintering state displayed a heterogeneous spatial distribution which was uncorrelated to *E. nitens* host tree physical characteristics. . Further work will examine temporal and inter-coup variation aspects by sampling with novel pitfall traps.

Acoustics and ecology – a study of the biology and acoustics of bush crickets in their natural habitat.

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The bush cricket *Leptophyes punctatissima* is a flightless tettigoniid that communicates using ultrasound. The acoustic behaviour of this species has been studied in the laboratory in the past. Experiments conducted in the field are providing information about distribution, migration, mate choice and acoustic behaviour, under natural conditions. Studies of the development of the immature stages suggest that there is a succession of food plants and that the choice of sites for egg-laying may be influenced by the food plant required by the first instars. Differences in preferences for food plants are known to exist between different populations and the implications of this have been studied in populations from different parts of the UK. Preference for food plants has been demonstrated to change during development from 1st instar stage to adult. Early instars show preference for species in the family Lamiaceae, in particular wood sage, *Teucrium canadense*. We suggest that plants in the Lamiaceae contain phytosterols essential precursor molecules for synthesis of cell membranes and steroid hormones. We discuss the type of environment favourable to this species of bush cricket, both from an acoustic and a developmental perspective.

Habitat loss decreases predator: prey ratios in a pine-bark beetle system

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Using a pine-bark beetle ecosystem as a model, we tested the hypothesis that the predator:prey ratio should decrease with increasing habitat loss at the landscape scale. We selected ten red pine (*Pinus resinosa*) sites, representing extremes of available habitat within a 2-km radius surrounding each stand (low: 14.9% vs. high: 55.1% conifer forest cover). The bark beetle, *Ips pini*, its predators and parasitoids were sampled using baited multiple funnel traps and naturally-infested trap logs. Effects of habitat loss were considerable; ratios of predators to prey (mean +/- SE) were significantly reduced in isolated stands (1.63 +/- 0.41) as compared to those with large amounts of surrounding conifer habitat (0.38 +/- 0.09). The decline in ratio occurred both because there was: 1) a lower abundance of predators (ca. 0.5-0.8 X), but also 2) a significantly higher number of prey (ca. 2.2 X) in isolated stands. Loss of habitat, therefore, differentially affected the two trophic levels, supporting predictions of theoretical models. Next, we investigate possible mechanisms causing higher prey populations in isolated sites: is this due to local predator-prey dynamics over successive generations or differential immigration, emigration, reproduction or mortality among the scolytid and its predators because of loss of habitat?

Fungal endophytes decrease aphid performance in grasses: effects on virus transmission?

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Systemic grass fungal endophytes may form a mutualistic relationship with their host plants. Endophytes live within the host tissues, receiving protection, nutrients, and transmission to the next plant generation via host seeds. The plant may benefit from endophyte infection through increased biomass production and improved herbivore resistance. Grass endophytes are able to produce toxic alkaloids, thereby negatively affecting several herbivore species. We examined how endophyte infection and nutrient availability affects the performance of bird-cherry aphid (*Rhopalosiphum padi*) in meadow fescue (*Lolium pratense*). Endophyte infection reduced aphid performance and the negative effects of endophyte infection were conditional on the soil nutrients; aphid performance decreased considerably on fertilized plants. This presumably indicates improved ability for investment in nitrogen-based alkaloid mycotoxins. The direct antiherbivore properties of endophytes can be exploited for instance in biocontrol. Barley yellow dwarf viruses (BYDVs) are aphid-transmitted viruses, which have worldwide distributions and infect species from the Poaceae. *R. padi* is an important vector species of BYDV. We study the interactions between BYDV, endophyte infection and aphids, and conduct experiments in field to find out whether endophyte infection inhibits virus transmission. In preliminary field experiments we have detected differences in virus incidences between endophyte-infected and uninfected *L. pratense*, indicating lower virus frequencies in endophyte-infected plants.

Melon: a new host of *Sesamia nonagrioides* **(Lefebre)**

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Sesamia nonagrioides (Lefebvre) (Lepidoptera Noctuidae) has been found for the first time on melon in a greenhouse in the Caserta province (South Italy) at the end of June 2003. Damage was caused by larval instars feeding on fruits and partly on leaves. Damage was observed on 70-80% of fruits. Eggs were laid on the leaves, from which newly hatched larvae moved to fruits, mostly boring at the bottom side. Moulting always took place in the fruits, where the head capsule of all instars were found. Larvae firstly eat tiny galleries, which are gradually enlarged up to 6-8 cm diameter. Even if erosions did not reach the flesh, fruits were seriously damaged because of invading larvae of Mediterranean fruit fly, *Ceratitis capitata* Wiedemann (Diptera), along with several saprophagi, as Coleoptera Nitidulidae (*Carpophilus* spp.), Diptera Phoridae (g. sp.) and Syrphidae Milesiinae (g. sp.). On growth completion, the fully fed larva crawled to the ground to pupate. Heavier losses occur when fruits are attacked in the last cycle phases, as they easily decay for pathogenic microorganisms (*Trichothecium roseum*, *Erwinia* spp.) growing on lesions produced by larvae. To kill adults before oviposition, pesticide applications, determined upon adult monitoring, are usually necessary.

Effects of life-history characteristics and resource predictability on population genetic structure of *Ogyris* (Lycaenidae) butterflies.

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Persistence of a butterfly metapopulation requires patches of host plants that are available for colonisation by dispersing individuals. The spatial and temporal predictability of patches is expected to influence the movement of individuals in the patch network. For example, a species utilising small, unpredictable patches may exhibit a relatively high level of dispersal in order to maintain the metapopulation. This study compares population genetic structure of the mtDNA COI gene in three *Ogyris* taxa (*O. amaryllis amaryllis*, *O. a. meridionalis*, *O. zosine*) from eastern Australia. The taxa differ in several life history characteristics that are hypothesised to influence patch predictability. The species with the most stable and predictable resource patches (*O. a. amaryllis*) displayed the strongest genetic structuring at both low and high clade levels. Species with less predictable resource patches (*O. a. meridionalis* and *O. zosine*) displayed less structure across clade levels. These results are consistent with the theory that spatial and temporal predictability of resources is an important influence on animal dispersal and population turnover.

Tri-trophic level interactions between the New Zealand flower thrips (*Thrips obscuratus*) and botrytis bunch rot (*Botrytis cinerea*) in grapes

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Botrytis Bunch Rot in grapes is caused by the fungus *Botrytis cinerea* and can lead to significant economic losses in vineyards world wide. The disease pathways of this fungus are numerous and complex, of which the spread by insects is the least investigated. The New Zealand Flower Thrips (NZFT), *Thrips obscuratus*, a thrips species endemic to New Zealand, is widespread in vineyards at flowering and thrips eggs, larvae, and adults can be abundant at that time and later, some can also be found in fruit bunches. Therefore, it seems likely that a large part of the life-cycle of NZFT may take place on the vines, inter-row plants and weeds that grow in, and in areas surrounding the vineyards. The first step to investigate the interactions between the fungus and the insect is to determine the population dynamics of the thrips in vineyards. At two field sites in the New Zealand South Island regions of Nelson and Canterbury, population monitoring of NZFT from spring to autumn was carried out. White water traps at three different heights were placed randomly in the vineyard and changed weekly. Additionally, 7.5m high suction traps (based on the Johnson-Taylor trap) were placed in those regions to monitor local flight activity. Results of the thrips population dynamics from those traps in grapes are given here.

Phenoloxidase-like activities and the function of virus-like particles in ovaries of the parthenogenetic parasitoid *Venturia canescens*

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The ichneumonid endoparasitoid *Venturia canescens* successfully develops inside the hemocoel of another insect by using maternal protein secretions, including nucleic acid-free virus-like particles (VLPs), to manipulate host physiology. These VLPs consist of four major proteins, which are produced mainly in the calyx tissue and transferred into the host insect together with the egg. One of the protein-coding genes (*vlp1*), with similarities to phospholipid-hydroperoxide glutathione peroxidases (PHGPx), exists in allelic forms producing two protein variants with different protein properties. We summarise observations indicating that oocytes and eggs are the source of reactive electrons, which potentially damage the lining and membranes of calyx tissues. We discuss the possible role of VLP1 in counteracting the damaging effects of oxidised phospholipids on membranes surrounding VLPs in the calyx lumen.

Lack of synchrony between apple leafcurling midge (*Dasineura mali*) and its parasitoid, *Platygaster demades*, limits biological control

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Apple leafcurling midge (ALCM, *Dasineura mali,* Kieffer) is found in all pipfruit growing regions of New Zealand. In central New Zealand there are five generations per year of which the second is the largest and most damaging to trees. The egg parasitoid, *Platygaster demades* (Walker) which was introduced to New Zealand in 1925 to control the pear leafcurling midge is the most important natural enemy of ALCM. However, despite high levels of parasitism, ALCM populations can still reach damaging proportions. Monitoring of ALCM and *P. demades* populations over three seasons on several Nelson orchards revealed differences in the synchrony of host and parasitoid generations. The parasitoid was largely absent when the second generation of *D. mali* began, allowing for a significant population build up. Percentage parasitism in later generations was high but population levels of *D. mali* were also high. Emergence data indicated that first emergence of *D. mali* adults from the first generation was 6-11 days earlier than for *P.demades*, thus allowing for most of the second generation midge eggs to escape parasitism. The influence of host and parasitoid phenology and parasitism levels on population control of ALCM is discussed.

Life history traits of the bamboo gall maker and its inquiline in the bamboo stand

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The developmental stages of the gall maker, *Aiolomorphus rhopaloides* Walker (Hymenoptera: Eurytomidae), and its inquiline, *Diomorus aiolomorphi* Kamijo (Hymenoptera: Torymidae), in bamboo galls were determined by examining soft-X ray photography in the stand of the bamboo *Phyllostachys heterocycla* Matsumura (Monocotyledoneae: Gramineae). The gall makers overwintered as pupae and its inquilines as larvae. Adults of the gall maker emerged from the galls from mid-April to early May, coinciding with the bamboo bud elongation period. This synchronization suggests that eggs laid in the new shoots have a nutritional advantage for resulting larvae, and that a slight advance or delay in emergence may reduce suitable oviposition sites, causing population fluctuations. The inquiline emerged from late April to early June during the bamboo shoot elongation period and females could lay eggs in elongated shoots providing relatively higher nutrition for larvae.

Adaptive significance of oviposition of diapause eggs in summer in the rice leaf bug, *Trigonotylus caelestialium*

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The rice leaf bug, *Trigonotylus caelestialium*, is one of the major insect pests of rice in northeastern Japan, because it intrudes into paddy fields and causes damage of pecky rice by sucking young grains. This species produces five or more generations a year in Hokuriku District and overwinters in the egg stage in diapause state. This egg diapause is maternally induced by short days at moderate temperatures in autumn. However, it has been reported that incidence of diapause eggs increases temporally in summer as well. In the present study, adaptive significance of oviposition of diapause eggs in summer in *T. caelestialium* was examined by field researches and laboratory experiments. Seasonal prevalence of diapause eggs was different in summer among research sites with different flora in an experimental field and it was shown that oviposition of diapause eggs follows deterioration of food conditions. In mid-summer, nymphal performance was worse because of food scarcity and thus oviposition of nondiapause eggs in summer seemed less adaptive. Therefore, oviposition of diapause eggs in summer is an adaptation for avoiding food scarcity and this indicates that *T. caelestialium* has a life cycle strategy with bet-hedging to survive the summer environments.

The influence of transmission modes, host wasp & host tree preference on the population structure of *Amylostereum* symbionts of siricid woodwasps.

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Siricid woodwasps (Hymenoptera) whose larvae develop in softwoods have an obligate symbiosis with species of the fungus *Amylostereum* (Basidiomycotina). While numerous Siricid species are known, only *A. areolatum*, *A. chailletii* and *A. laevigatum* are known to have such specific symbioses. The association is species specific for the wasp, but not for the fungus. Two wasp species carrying the same fungal species often prefer different host trees to oviposit in. For example, *S. juvencus* and *S. noctilio* overlap in regional occurrence and both carry *A. areolatum*, but the former species preferentially breed in *Picea*, while the latter prefers *Pinus*. Here we study we the consequences of wasp association and tree host on population structures of *Amylostereum* spp. We also compare populations from different geographic areas in order to characterize patterns of spread of both symbiotic partners. Transmission of the fungus between generations is thought to be vertical (i.e. from mother to daughter). The shared fungal species between different wasp species, and oviposition of wasps in the vicinity of previous Siricid and *Amylostereum* infections, however, indicates a possibility for horizontal transfer as well. Using population and phylogenetic data of the fungus we aim to evaluate the influence of different modes of transmission.

Impacts of perchlorate and hexavalent chromium at the bottom of the food web

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Perchlorate and hexavalent chromium (Cr VI) are significant anthropogenic pollutants in many areas of the world. We investigated the individual and joint effects of these pollutants on the bottom of the food web in representative ecosystems. The primary test organisms were *Megaselia scalaris*, a terrestrial detritovore, and *Culex quinqufaciatus*, a common vector of encephalitis. For *M. scalaris*, Cr VI slowed development and acted as a toxicant. However, perchlorate increased the rate of growth and was relatively non-toxic. In combination, the joint effects canceled, and development was not significantly different from untreated controls. Thus, removal of either pollutant will have substantial effects on the fitness and survival of these insects. We also tested the hypothesis that perchlorate, Cr VI, or the combination of pollutants, would interact with control strategies for mosquito larvae based on the bacteria *Bacillus thuringiensis var israelensis* (Bti). Significantly more Bti (57%) was required for control when perchlorate was removed from the system. The implications of this research for both ecosystem health and mosquito control are substantial.

The population dynamics of genetically modified mosquito populations in laboratory microcosms

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Over recent years, rapid progress has been made in the stable germline transformation of mosquitoes. The ultimate aim of this technology is the mass release of transgenic mosquitoes into the field but there is a paramount pre-requisite to determine the potential risks and environmental consequences associated with such a release. Fundamental to any risk assessment is the need for a full understanding of the life history characteristics of the transgenic individuals and populations in relation to wild-type mosquitoes. Any effect of genetic modification on the reproductive capability or competitive ability of the transgenic mosquito will necessarily have an impact on the dynamics of the population as a whole and, ultimately, it's ability to invade and become established in natural populations. The approach which we have adopted is to explore the population ecology of transformed mosquitoes in laboratory microcosms. We have established continuous "free-flow" cultures of *Aedes aegypti* mosquitoes, stably transformed with the EGFP gene. We are able to monitor and analyse the patterns in abundance of all developmental stages of transformed and non-transformed mosquitoes over multiple generations. Furthermore, the population dynamics are underpinned by experiments that explore the impact of competition for resources on measures of fitness such as survivorship, developmental rates and fecundity.

Dispersal of rice leaf bug, *Trigonotylus caelestialium,* in Italian ryegrass fields

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The rice leaf bug, *Trigonotylus caelestialium* (Kirkaldy), is known as a pest which attacks developing rice grains and causes pecky rice. To trace the movement of the adult bugs, mark and recapture experiments were carried out in Italian ryegrass fields. For labeling the adults, rubidium was used as an internal physiological marker. Field collected adults were labeled and released to field plots where Italian ryegrass was cut at different date. Sampling censuses with a sweep net were conducted within the released plot at 1 and 2 days after release. The population parameters were estimated using Yamamura's method (Yamamura, 2003), on the assumption that the probability of capturing the bugs was constant during two consecutive sampling censuses. The estimated rates of colonization were 0.44 - 1.0 for female and 0.33 - 0.71 for male, respectively. The results showed that females are less likely to disperse than males.

Intensity of density effects in larval performance and oviposition pattern in drosophilid flies

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Spatial aggregation of eggs has been suggested to promote coexistence of competing species on patchy, ephemeral resources. In former studies, however, aggregation was assessed by species composition and abundance of emerging adults from patches instead of the egg distribution. This might produce a bias in the assessment of aggregation if interspecific relationship, for example, superiority in competition, differs among species or changes density-dependently. Additionally, if the effect of density on performance or survival differs among species, oviposition pattern may also differ among them; for example, species that receives strong negative effect at high density may distribute eggs uniformly. Here I examined 1) the effect of density on performance and survival in five drosophilid species, 2) interspecific relationship (e.g., competitive or mutualistic relationship) among them, and 3) patterns of egg distributions in them. It appeared that the effect of density varied among species. In addition, the outcome of competition differed with density, suggesting the inappropriateness of the assessment of egg distribution by emerging number of adults. Species showing high performance even at high density tended to exhibit highly aggregated egg distribution. The connection between the intensity of density effect and oviposition pattern would affect coexistence of them.

Causes of pest outbreaks

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Contrary to a well-known opinion, biodiversity in arable fields may be higher than in natural adjacent biotopes and its level has no relation to pest outbreaks in crops. The outbreaks arise due to: 1) the monoculture crop itself is a strong source of olfactory and visual attractive stimuli for herbivores but not always for predators and parasites colonising the field later when there is sufficient amount of their victims there; 2) many crop pests are hortobionts and can be invulnerable because many hortobiont predators are able to penetrate into the field only after "canopy forming" by the plants. Herpetobiont predators climb plants relatively seldom; 3) the most of entomophagous species prefer the edge of the fields, hence the central zone is more favourable for pests; 4) almost all natural enemies use field grassy margins as shelters for overwintering and in other unfavourable situations. These narrow strips of land is often insufficient to supply all territory of the field with entomophagous arthropods; 5) some herbivores increase their populations fast (*i.e.*, aphids) and their natural enemies are often not able to overtake them.

Shoot sprouting of three willow species in response to damage by a stem-boring insect affects insect herbivores at different intensities

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We investigated the indirect effects of the stem-boring insect *Endoclyta excrescens* on two insect herbivores on three willow species, *Salix gilgiana, S. eriocarpa*, and *S. serissaefolia.* When the branches were damaged by the stem-boring insect, the willows were stimulated to vigorously produce lateral shoots. This enhanced lateral shoot growth was also found after physical damage by artificial boring. Newly-emerged lateral shoots were longer and upper leaves had a higher water and nitrogen content. Larvae and adults of the leaf beetle *Plagiodera versicolora* were significantly more abundant on lateral shoots than on current-year shoots. Similarly the density of the aphid *Chaitophorus saliniger* was significantly higher on lateral shoots than on current-year shoots. Although densities of the two insect species on current-year shoots did not differ among willow species, we found significant differences in densities on lateral shoots among willow species. The stem-boring insect positively affected the aphids and the leaf beetles by producing new food resources as a result of the resprouting responses of the three willow species. However the intensity of the positive effects caused by the stem-boring insect was different among the three willow species because of different regrowth responses to boring damage.

Arthropods living in the subnivean space in the Dolomites (north-eastern Italy)

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In the temperate regions, during the winter, snow cover forms an insulating blanket between two very different environments: the y, very cold and windy atmospheric air environment and the moist, relatively warm and stable-air microenvironment under the snow. In the subnivean space (3-5 cm between snow and soil) the high humidity, constant oxygen input and temperatures near 0°C allow some invertebrates to be active. During two consecutive winter seasons, we collected arthropods active under the snow cover using modified pitfall traps in a valley of the Dolomites (NE Alps). Arachnida (12%), Collembola (80%), and Insecta (18%) were found. The most abundant insects were Coleoptera (39%) and Diptera (43%). Some species of Collembola and Diptera, found active under the snow, are known to be troglophilous: specimens of *Chionea* have been reported during the summer from natural and artificial cavities, whereas the collembolan species *Tomocerus minor* and *Arrhopalites caecus* had so far often been collected from caves. The presence of troglophilous species shows that the subnivean space becomes an expansion of underground microhabitats, as they are both characterized by constant microclimatic conditions.

Biological assessment of non-wadeable Midwestern (USA) rivers using macroinvertebrates

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There are relatively few state or federal programs designed to monitor and evaluate non-wadeable rivers. Many of the more traditional population and community level metrics used in wadeable stream assessment may not be appropriate for assessing non-wadeable systems. We analyzed macroinvertebrate samples from non-wadeable river reaches subjected to a wide array of anthropogenic influences to determine which potential metrics provided the most unique information and accounted for the largest amount of among-reach variability. The final protocol allows for two types of assessments: One that targets large woody debris habitats only, and one that relies on samples from all habitats present in the study reach. Both assessment types evaluate sites based on scores from 7 individual metrics incorporated into a summary index. When properly implemented and combined with the habitat assessment procedures, the macroinvertebrate protocol will allow for the objective evaluation of non-wadeable rivers in the midwestern United States and may provide a framework for evaluating non-wadeable river systems in other regions.

Modelling population dynamics of light brown apple moth *Epiphyas postvittana* (Walk.) (Lepidoptera: Tortriciade) in Australia: how many generations are there?

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The light brown apple moth (LBAM) is a serious pest of fruit crops and ornamentals. It is native to southeast Australia including Tasmania and has been introduced to New Caledonia, British Isles, Hawaii, Western Australia and New Zealand. The pest is highly polyphagous, causes significant economic damage to its principal host apple and has been classified as a noxious pest in USA and Canada leading to restrictions on imports of Australian primary produce. The moth is cold adapted and believed to undergo three generations a year including a summer, autumn-winter and spring generations in Victoria, in the Australian Capital Territory, in Tasmania and in New Zealand. Knowledge of the biology and ecology of this species is relatively comprehensive and the published data have been re-analyzed and used in the construction of a population dynamics model. The model is built using DYMEX^{TM}, a process based modular modelling package for constructing stage structured population models. The model is designed to be driven by the climate data. The model predictions indicate the three generation phenology with climate data from locations like Victoria and Hobart (Tasmania) but 4 generations in locations like Mildura and Victoria. These predictions lead us to re-assess the estimated number of generations previously reported for LBAM.

Can volunteers be used to estimate the abundance & diversity of butterflies using the Pollard walk? An Australian example.

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In some circumstances volunteers can be used to collect various ecological data. We have been investigating the usefulness of volunteers in collecting data on the abundance and diversity of the urban butterfly fauna in Adelaide, South Australia. The program is modelled on other programs such as the British butterfly monitoring scheme. Volunteers received training to record the diversity and abundance of butterflies using a 'Pollard walk' protocol. Kits containing information on butterfly identification, data sheets, and instructions for conducting surveys were provided to each volunteer. We analysed the effects of factors such as weather conditions, observer experience, and whether or not observations were made on an individual or group basis. Our preliminary results indicate that volunteers are useful in detecting and identifying butterflies of certain groups *e.g.* Nymphalidae, Papilionidae, but less efficient in detecting and identifying other groups of butterflies, *e.g.* Hesperiidae and Lycaenidae. This may be due, in part, to the similar colouration and cryptic behaviour of butterflies from these families. Unfortunately, the latter two groups contain several taxa of conservation concern and therefore this censusing technique may have some shortcomings in identifying conservation priorities for butterflies

Estimation of leafhopper biodiversity (Auchenorrhyncha: Cicadellidae)

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Leafhoppers are an important component of terrestrial food web, because they occur in high density and species richness in most terrestrial ecosystems. To assess true species richness of leafhoppers is no easy task, because extraordinarily intensive field surveys are needed. When leafhopper diversity is surveyed by an intensive but short-term field survey, it is required to explain that it is true diversity. In this study, the author tried to indicate the most suitable estimator to estimate true species richness for leafhoppers using statistical sampling data. Although most of the estimators such as Chao1 were not consistent with the true species richness, an incidence-based coverage estimator of species richness was the most appropriate method to estimate the true species richness. The estimator, ICE, estimates the total species number of leafhopper in the sensus field, but it does not show each component species of fauna. To estimate component species, possibility of an occurrence for each species is simulated using floral data and GIS meteorological data without using the aforementioned estimator, because all of leafhoppers are phytophagous insects and their occurrence strongly depends on the existence of their host plants and meteorological elements.

Antipodes antipasto: mice make a meal of subantarctic insects

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The introduction of mammalian predators to New Zealand has had a major detrimental impact on the native fauna. Predation by introduced mammals has caused the extinction of many species and reduced the numbers of numerous others. Most research, however, has focused on the impacts of larger predator species (cats, mustelids and rats) on the avifauna. Invertebrate survey work on the subantarctic Antipodes Islands, 730 km southeast of New Zealand, shows the negative impact of the introduced house mouse (*Mus musculus* Linnaeus) on the invertebrate fauna. Comparisons between mouse-infested Antipodes Island, where mice were introduced about 100 years ago, and neighbouring, mouse-free Bollons Island indicate that mice have had a significant impact on the faunal composition and abundance of Antipodes Island invertebrates. Two species, *Loxomerus* n. sp. (Coleoptera: Carabidae) and an undescribed weta (Orthoptera: Rhaphidophoridae), may be extinct on Antipodes Island, probably due to mouse predation. Evidence also strongly indicates that mouse predation has markedly reduced the abundance of most of the larger beetle species (length > 4 mm). Long-term impacts of mouse predation and prospects for mouse eradication are discussed.

Comparison of the weevil settlements (Coleoptera: Curculionoidea) of four reservoirs in central Serbia

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The weevils investigations in Kragujevac and surroundings were initiated in 1987. Parallel study of four Reservoirs (Gruža, Grošnica, Šumarice and Bubanj) started in 2001. All representative habitats, from the lakes shoals to 50-100m from the water were covered: swampy meadows, moist meadows, valley meadows, neglected lands, ruderal vegetation, boundaries and tilled grounds, as well as forests. The sampling techniques included sweeping and shaking of plants, rinsing of aquatic plants, beating of the tree branches and bushes, mowing of the ground floor vegetation, hand collecting and separation of insects from the soil with the aid of Tulgren-Berlesse apparatus. Comparatively presented results are:

Reservoir	Findings	Species	Males	Females	Specimens
Grošničko lake	181	98	159	145	304
Lake in Šumarice	440	143	451	487	938
Bubanj lake	73	40	70	85	155
Gružansko lake	160	69	219	246	465
Total	854	207	899	963	1862

The focus was on aquatic weevils. During this study most of this kind of the weevils (like *Bagous bagdatensis* Pic) is registered for the first time in Serbia.

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Similarity of the soil weevil fauna (Coleoptera: Curculionoidea) in the west, central and east Serbia

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According to the authoress weevil materials collected from the soil since 1980s, on different localities in Serbia (West: Suvobor, Čemerno, Mučanj, Ivanjica; Central: Gledićke planine, Gruža, Rudnik, Bukulja, Žeželj, Grošničko jezero, Jastrebac, Goč; East: Beljanica, Kučajske planine, Resavska klisura) Central Serbia is a real bridge between West and East Serbia. There are 12 species. Six of them are common in the West (*Ruteria hypocrita, Acalles camelus, A. commutatus, A. echinatus, Acallocrates colonnelli* Bahr, 2003 and *Aparopion costatum*), and five in East Serbia (*Echinodera valida, E. behnei, Acalles papei, Aparopion costatum* and *Ubychia holdhausi*). Just one Cryptorhynchinae species, *Acalles ganglbaueri* (on Goč mt.), and quite common in the oak forests short-nosed *Brachysomus hirtus* are registered only in Central Serbia. *Aparopion costatum* is the only one soil weevil species common for all. *Acalles micros* has up to now been collected only in the East Serbia (on Beljanica mt.). The similarity expressed through the Sørenson index is: West-Central 0.667, Central-East 0.556 and only 0.167 for West-East.

How does cultivation in the vineyard affect beneficial invertebrates?

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Cultivation of the soil has been a common management practice in Australian vineyards for many decades. This is especially true for the irrigated vineyards in the Sunraysia region of north-west Victoria. Growers tend to cultivate for a variety of reasons including, frost protection, decreased soil compaction and increased drainage. However, cultivation also tends to disturb the soil, thus disrupting soil invertebrates such as earthworms and carabid beetles. Invertebrates are known to react sensitively to viticultural management practices and are considered important for their bioindicative value. An investigation into the impact of cultivation on soil and canopy invertebrates in vineyards in Sunraysia is in progress as part of my PhD. The experimental design of this study consists of five randomised plots within one vineyard. Each plot contains cultivated and non-cultivated areas. Within each plot, samples are collected along a transect using pitfall and yellow stick traps. Samples are being sorted to order, where analysis will determine which orders are significantly affected by cultivation. These orders will then be sorted to species level. In this experiment potential indicators of cultivation impacts will be identified and tested further in a regional study set to begin later this year.

Coleoptera in agrolandscape of Kuban-Azov lowland.

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Coleoptera have been studied in agricultural fields at different distances from their edges and in adjoining biotopes (in forestbelts, field grassy margins) for 5 years. About 1000 species of 69 families were found in this landscape seriously affected by man activity. Maximal numbers of Coleoptera were registered in field grass margins and in edges of the fields. In the central zone of fields, number of species is considerably reduced. The following families had the maximal number of species: Curculionidae (123), Staphylinidae (107), Chrysomelidae (104), Carabidae (71), Cerambycidae (42), Scarabaeidae (37), Tenebrionidae (36), Buprestidae (32), Elateridae (31). Some species collected in lowland fields were typical for the mountain meadows and woods, i.e. some representatives of Staphylinidae, Scaphidiidae, Trogositidae, Cucujidae, Erotylidae and Curculionidae. These beetles were founded on the plain only in spring.

Omaliopsis (Coleoptera: Staphylinidae) – old or new south-north connections?

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At least 16 lineages of the vast family Staphylinidae have endemic species disjunct among three or more austral areas, usually Australia/New Zealand/southern South America. The only lineage analyzed to date appears to have a New Zealand clade as sister to the rest of its species, which show multiple connections between South America and Australia. The genus *Omaliopsis* currently includes six African or Madagascan and two Chilean species; it was originally characterized as one of several "old Gondwanan lineages" of Omaliinae. I have discovered that *Omaliopsis* includes 40 more species (mostly undescribed) adding to its distribution northeastern India, Australia, the northern Andes, New Guinea, and non-Gondwanan areas Taiwan, southern China, Borneo, and Mexico (to southwestern USA). The sparse fossil record shows that Staphylinidae, perhaps even Omaliinae in particular, were present and widespread on Pangaea. Phylogenetic relationships within a widespread and potentially ancient clade such as *Omaliopsis* could be expected to reflect, at least partly, Pangaean or Gondwanan-Laurasian fragmentation. Reconstruction of relationships within *Omaliopsis*, with its northern and southern elements, will allow testing the origins of the distribution. In particular, the phylogenetic position of the southern Asian species differs in the predictions made by Pangaean and Gondwanan breakup models.

Modelling population dynamics of light brown apple moth *Epiphyas postvittana* (Walk.) (Lepidoptera: Tortriciade) in Australia: how many generations are there?

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The light brown apple moth (LBAM) is a serious pest of fruit crops and ornamentals. It is native to southeast Australia including Tasmania and has been introduced to New Caledonia, British Isles, Hawaii, Western Australia and New Zealand. The pest is highly polyphagous, causes significant economic damage to its principal host apple and has been classified as a noxious pest in USA and Canada leading to restrictions on imports of Australian primary produce. The moth is cold adapted and believed to undergo three generations a year including a summer, autumn-winter and spring generations in Victoria, in the Australian Capital Territory, in Tasmania and in New Zealand. Knowledge of the biology and ecology of this species is relatively comprehensive and the published data have been re-analyzed and used in the construction of a population dynamics model. The model is built using DYMEX^{TM}, a process based modular modelling package for constructing stage structured population models. The model is designed to be driven by the climate data. The model predictions indicate the three generation phenology with climate data from locations like Victoria and Hobart (Tasmania) but 4 generations in locations like Mildura and Victoria. These predictions lead us to re-assess the estimated number of generations previously reported for LBAM.

Fires, forest mosaics & boreal beetle diversity

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Forest composition, age structure, density, and growth rate vary in space, providing a range of different habitats for boreal wildlife. Carabid beetle (Coleoptera: Carabidae) communities are sensitive to habitat variation and, therefore, we studied concomitant spatial variation between forest habitat and carabid beetle communities. A systematic grid of 200 plots (640 m apart) was established on the EMEND study site (7 km by 13 km of boreal mixedwood in northwestern Alberta). Forest characteristics and beetle assemblages collected by pitfall trapping were compared in each of these 200 plots. Distinct late successional tree and beetle species assemblages occurred mainly on north facing aspects of the landscape where fires have been uncommon during the last 150 years.

Bugs beneath the surface: biodiversity, degradation & the functional roles of soil macroinvertebrates in maintaining ecosystem health in Australia's tropical savannas

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Soil macroinvertebrates such as termites and ants play a pivotal role in sustaining production and biodiversity in Australia's tropical savannas. Through their foraging and nesting activities they recycle nutrients and carbon and produce soil pores that facilitate water infiltration. The challenge ahead is to quantitatively understand the relationships and process that drive this. What roles do different species and functional groups play in various landscape processes? What are the effects of disturbances (eg. cattle grazing) on these relationships, and the consequences for landscape health? Increased degradation is often associated with declines in macroinvertebrate diversity and activity. Termites appear to be one of the most resilient groups, capable of maintaining activity in degrading landscapes. This study examines the effects of long-term stock exclusion on termite abundance, diversity and function in Queensland, and the use of termites as a tool to kick-start the rehabilitation process in degraded savanna sites in the Northern Territory. This paper presents (i) an overview of the importance of soil macroinvertebrates to soil functioning, and (ii) experimental results that suggest variation in macroinvertebrate community structure and soil water relations with changing land condition, and the role of termites in soil rehabilitation.

Canopy beetles of the western Amazon Basin: β-diversity in time and space

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Beetles sampled as part of a monitoring program of Oil Company activities in the western Amazon Basin were subsequently reinvestigated to test for β-diversity between two western Amazon rainforest sites located on the equator at 21 kilometers distance from each other. Coleopteran families selected for this study were Artematopodidae, Attalabidae, Buprestidae, Cantheridae, Carabidae, Ceratocanthidae, Chrysomelidae, Cleridae, Curculionidae, Elateridae, Erotylidae, Mordellidae, and Tenebrionidae. A total of 14,622 adults representing 1915 morphospecies were processed from 1200 samples (10,944 m^2 trap surface area). Assessments using EstimateS$^{\copyright}$ were made to determine whether the sampling regime was adequate to obtain a complete representation of species in the two plots. For those families adequately sampled, a test of Complementarity for the two plots resulted in values ranging from .48 to .69, the species-rich families having the higher values. These results suggest a markedly high degree of species turnover across small distances in generally similar forest composition in the western Amazon Basin, which in turn suggests that a conservation strategy of making a few large conservation areas may not preserve a large share of existing biodiversity. This further suggests that a new conservation paradigm is necessary if we are to protect much of Earth’s precious natural heritage.

The identification and ecological role of dipteran larvae in tertiary fossil lake ecosystems

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The sediments of fossil lakes often yield numerous remains of terrestrial insects. In contrast to this, in many fossil sites autochthonous, water-living forms are only rarely found. This is the case for the former Maar lakes of Messel, Eckfeld and Enspel (all Germany). Thus, analysis of the insect fauna may yield interesting clues to the terrestrial environment of the former lake but is not able to yield information on the ecosystem of the lake itself. Our analysis of fossil faeces from planctivorous fish of the fossil site Messel has for the first time revealed that larvae of phantom midges (Chaoboridae) and biting midges (Culicidae) existed in the former lake. The identification of the larval midges was possible on the base of isolated mouthparts, i.e. mandibles and filter tufts. Due to the quantity of these remains it can be concluded that the aquatic midges formed large populations in the former lake. Therefore, the larvae and to a lesser degree also the pupae of these midges were very important factors of the food web and are crucial elements for the understanding of the function of the former lake ecosystem.

From metapopulation patterns to processes at the individual level: lessons from long-term butterfly studies

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Metapopulation biology provides a sound theoretical framework for the understanding of population persistence in fragmented landscapes. Long-term comparative studies of metapopulations of several butterfly species in contrasted landscapes generate some results that might be relevant for the future of research in the field: (1) dispersal rates decrease according to the fragmentation process, which leads to dispersal rates adapted to each metapopulation in its particular habitat network and (2) local population growth rates are related to the rate of efficient dispersal. Two related processes acting at the individual level may generate such patterns: a decrease of the dispersal propensity due to higher dispersal mortality according to fragmentation, which subsequently modifies the trade-off between dispersal and fecundity. The first process linking lower dispersal propensity with higher cost of dispersal is documented by solid empirical evidences, whereas the change in the trade-off between dispersal and fecundity is currently inferred from interspecific comparisons only. According to both processes, selection against dispersal in fragmented landscapes would lead to changes in individual reproductive performances, which could affect the evolutionary future of the population. From a conservation perspective, individuals from (meta)populations and species sacrificing dispersal at the expense of reproductive performances are most vulnerable to habitat loss and fragmentation, because of their higher sensitivity to stochastic events.

Indirect interactions and quantitative food webs

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Quantitative food webs are a means by which the multitrophic structure of natural communities can be described, and hypotheses about indirect interactions formulated. We will describe the construction of quantitative food webs representing communities of plants, herbivorous insects, parasitoids (and hyperparasitoids), in particular communities based around aphids and leaf miners. We will show how the webs can be used to predict possible examples of apparent competition, and how these hypotheses can be tested using experimental manipulations in the field.

Trait-mediated indirect interactions between herbivorous mammals and insects: claiming for an integrate view of the terrestrial communities

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My goal in this presentation is to assess the importance that the trait-mediated indirect interactions (TMII) play in the interaction between herbivorous mammals and insects, by reviewing the existing literature on both the diverse effects of mammals on plants and on competition between phytophagous insects and mammals, and by showing my own studies on mammal-insect-crucifer interactions. The main idea is that TMIIs are crucial for insects since in most cases 1) ungulates outcompete insects, and 2) the main short-term effect of ungulates is not a change in plant density but a modification of chemical, physiological, morphological and architectural traits of plants (induced resistance). I will also extend the consideration of TMIIs to those phytophagous insects like nectar- or pollen-feeders that are mutualists of plants, specifically when both ungulates and insects select plants according to the same traits. In addition to the potential ecological consequences of TMIIs, I will argue that ungulates and insects can sometimes act as conflicting selective pressures on plants, since an evolutionary consequence of TMIIs has been little considered. All of these ideas suggest the necessity of considering together both phytophagous vertebrates and invertebrates in order to draw a more accurate picture of the dynamics of terrestrial communities.

Climate change and recovery of endangered species

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In a quantitative analysis of 99 wild species, Parmesan and Yohe (2003) estimated that the magnitude of range shifts with 20th c. climate warming was 6.1 km/decade poleward. Because endangered species typically live in rare habitats, either historically or as a result of habitat destruction, shifts of this magnitude in climate space will have profound implications for the abilities of endangered species to persist in traditional preserve systems. Under climate change, management and recovery of endangered species will necessitate a fundamentally different approach from traditional recovery plans. These processes (climate change impacts and management options) will be discussed in light of an endangered sub-species of butterfly, the Quino Checkerspot (Euphydryas editha quino), which occurs in southern California, USA.

Dynamics of insect-plant association in a human-disturbed butterfly metapopulation

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A relatively homogeneous patch network for Edith's checkerspot butterfly (Euphydryas editha) was converted by humans to a network with two patch types: forest clearings and unlogged patches (rocky outcrops). In the clearings a novel host species, Collinsia torreyi, was colonized. Initially, clearings supported higher population growth than outcrops and acted ecologically as a set of sources. Metapopulation-level effects that resulted from this role included effects of isolation from clearings on both insect density and host preference in outcrops. Evolution of host preference in outcrop patches was driven by gene flow against the direction of natural selection in those patches. Ecological stability of the overall system was augmented by a negative association between the habitat quality of clearings and the quality of adjacent outcrops. High quality clearings lay adjacent to low quality outcrops and vice versa. For about 20 years the system comprised a reversible source-sink with alternative stable states. Clearings could act either as sources or as sinks. With succession the reversibility of the source-sink system has evaporated and the patch type that comprised strong population sources in the 1980's, the clearings, now consists of a set of permanent sinks. The insects have all returned to their traditional host species. Nonetheless, a strong evolutionary legacy of the events of the past 20 years can still be detected in the system.

Assessing predation rate of biological control agents using video techniques

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In biological control research, key questions include: Which are the most important predators or parasitoids in a particular agro-ecosystem? What host/prey species do they attack? For particular or parasitoid species, a further question is: What is the attack rate or prey/host consumption rate? Answering the first two questions above can involve assessing natural enemy density, followed by dissection and/or immunological or molecular analyses of diet. The third question is particularly difficult to answer in the field. Real-time observation of the natural enemies in the field is possible but is very time consuming. The use of "prey facsimiles such as laboratory-produced blow fly eggs or Drosophila pupae can give "predation rate" but, alone, does not identify the predator species. Using prey facsimiles or real prey (in combination with infra-red illumination for nocturnal predators) along with time-lapse video recording can address this key question more easily. Analogue video recording using tapes has many drawbacks including tape and VCR wear and time-consuming searching for relevant parts of the recording. Digital video can overcome this problem. The potential of this method is illustrated with examples from nocturnal polyphagous predators on the soil surface and from parasitoid f

Competitive coexistence in patchy environments: the interplay between spatial variation and density-dependent dispersal

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It is well known that dispersal from localities favorable to a species' growth and reproduction (sources) can prevent competitive exclusion in unfavorable localities (sinks). What is perhaps less well known is that too much emigration can undermine the viability of sources and cause regional competitive exclusion. I investigate two biological mechanisms that reduce the cost of dispersal to source communities. The first involves increasing spatial variation in the strength of competition such that sources can withstand high rates of emigration; the second involves reducing emigration from sources via density-dependent dispersal. I compare how different forms of spatial variation and modes of dispersal influence source viability, and hence source-sink coexistence, under dominance and preemptive competition. A key finding is that while spatial variation substantially reduces dispersal costs under both types of competition, density-dependent dispersal does so only under dominance competition. Thus, the diversity reducing effects of high dispersal rates persist under preemptive competition even when dispersal is density-dependent, but can be significantly mitigated under dominance competition if density-dependent dispersal occurs in response to crowding rather than Allee effects. These results lead to testable predictions about source-sink coexistence under different regimes of competition, spatial variation and dispersal.

Diversity-stability relationships in host-parasitoid communities: an empirical exploration

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The relationship between diversity and stability is crucial in understanding the dynamics of multi-trophic interactions. There are two hypotheses about the causal link between diversity and stability. The first is that fluctuations in resource abundance allow consumer coexistence, thus increasing diversity at the consumer trophic level (resource variability hypothesis). The second is that interactions between coexisting consumer species reduce consumer efficiency and damp consumer-resource fluctuations, thus increasing stability (consumer efficiency hypothesis). The two hypotheses lead to three predictions: (i) Fluctuations should be greater (resource variability) or smaller (consumer efficiency) in resource populations with coexisting consumer species, compared to those invaded only by the consumer species superior at resource exploitation, (ii) Average resource abundance should be greater (resource variability) or smaller (consumer efficiency) in resource populations with greater fluctuations, and (iii) Removal of the inferior resource exploiter should increase or unaffect resource population fluctuations (resource variability), or increase them always (consumer efficiency). I tested these predictions with data from a host-multiparasitoid community. The results contradict the expectations of the resource variability hypothesis, suggesting that host population fluctuations are unlikely to be driving parasitoid coexistence. They are consistent with the consumer efficiency hypothesis, that interactions between coexisting parasitoid species damp host fluctuations.

Estimation of developmental zero and thermal constant for alfalfa root weevil ((Sitona discoideus Gyll.(Col.: Curculionidae)) egg

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The alfalfa root weevil, Sitona discoideus Gyll, is one of the important pests of lucerne. This insect have wide distribution in Iran, and cause loss of yield and longevity of lucerne fields. The knowledge of developmental zero and thermal constant is a method that will be effective for estimation of time of activity and increasing accuracy of control methods. The eggs were reared in five constant temperatures (15,20,23,26, 28 &30°c) and 100% R.H., and developmental rates were calculated. With using of five models, Linear, Logan, Logan6/Lactin1, Lactin2 and Biere2, evaluated relationship between constant temperatures and developmental rates. The linear model had the most efficiency. This model estimated developmental zero and thermal constant.

Spatial distribution of immature stages of alfalfa seed weevil, Tychius aureolus(Col.: Curculionidae), and alfalfa seed wasp, Brochophagus roddi, (Hym.: Eurytomidae) in alfalfa seed fields

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Alfalfa seed pests, such as alfalfa seed wasp, Brochophagus roddi (Hym.Eurytomidae) and alfalfa seed weevil, Tychius aureolus (Col.Curculionidae), cause high damage in alfalfa seed production, but do not development any sampling program for population estimate and management of them. In during two years (2001& 2002), the spatial distribution of immature stages of these insects was analyzed using the crowding indices (ID ,Cx ,X*, Id & 1/k) , Taylor's power law and Iwao's regression technique. All indices indicated an aggregated distribution of two insects in lucerne seed fields. Compared with Iwao's regression, Taylor's power law provided a better description between variance and mean density. The coefficient of Taylor's power law for alfalfa seed wasp and alfalfa seed weevil were (a= 0.5, b= 1.02 ± 0.06, r2 = 0.95) and (a= 0.275, b= 1.06± 0.05 r2= 0.82) resp. and these for Iwao's regression were (a= 0.8, ß= 2.95± 0.06 r2= 0.5) and (a= 0.9, ß= 1.07± 0.14 r2= 0.29) resp.

Biodiversity, an emergent property of plant-mediated indirect interactions?

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Although, indirect interactions among species, particularly herbivores, have been considered unimportant, most terrestrial ecosystems lose 15-20% of above-ground net primary productivity (ANPP) to herbivores. There are at least 4 major plant-mediated pathways through which herbivores can indirectly impact biodiversity of dependent community members. Individually, specific herbivores can alter plant architecture, plant secondary chemistry, plant species composition, and/or build structures all of which can affect microclimate, host-plant quality, and host-plant spatial distribution and abundance. In natural systems species rarely exist in isolation, but are part of a greater tangled web of direct and indirect effects of many species as well as biotic and abiotic ecological factors. Because of this tangled web of interactions, synergistic community responses (i.e., responses that are attributable to a combination of factors rather than any single factor alone) are probably common, though rarely studied. In an aspen and cottonwood system I show that arthropod species diversity is driven by the synergistic impact of plant mediated indirect interactions among common herbivores, including a gall-forming sawfly, elk, and beavers. Specifically I argue that the individual and combined effects of common, dominant and keystone herbivores create a gradient of habitats or conditions that increase the probability of unique indirect interactions, increase the heterogeneity of potential niches for other species, and increase, the possibility of synergistic outcomes, all of which can positively affect biodiversity.

Integrating mark-recapture, weather, and remote sensed data to predict movement

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The most difficult part of understanding population dynamics is acquiring movement data that can be analyzed in a sound theoretical context. The movement of animals among resources has been studied using many empirical techniques and analytic theory. I will use arthropod systems to show technical challenges and analytic methods that may be applied to your system. I show how multiple approaches may provide a synthesis for understanding movement in a given system, and I finish by couching the results in simulation studies that predict population dynamics.

Metapopulation dynamics of host-parasitoid interactions: the role of space and stochasticity

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The pairwise interaction between parasitoids and their hosts has shaped our understanding of fundamental and applied concepts in ecology and in particular in population dynamics. Recently, work to extend these concepts to more spatially-explicit interactions has revealed how patterns of persistence in host-parasitoid systems may be influenced by both ecological and stochastic factors. In this talk, we will focus on how the patterns of host-parasitoid interactions are altered in spatially-explicit environments using both quantitative methods and experiments. In particular, we will focus on the use of a time series approach to understanding how space can affect persistence and how stochasticity can affect the dynamics of specific host-parasitoid interactions. We will briefly discuss the sorts of methods we have used to develop our understanding of these host-parasitoid systems.

Coexistence and niche segregation in cockroach guilds in cultivated fields

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Cockroach guilds were surveyed to investigate the coexistence and niche segregation of different species. Habitat selection of each species was described in relation to environmental factors. Guilds of seven species present in sugar-cane fields on La Réunion Island were studied for one annual cycle in fields with different ecological parameters. Structure variations of cockroach communities were analysed at the field scale, at the sample unit scale and according to variations related to the cycle of sugar-cane growth. A Canonical Correspondence Analysis was used to elucidate relationships between species diversity, population abundance and environmental characteristics. Our results allowed us to characterize the ecological niches of these cockroach species and to explain the specific patterns of their establishment and population outburst in sugar-cane fields. Three pairs of species were evidenced, each pair occupying a particular type of habitat. The seventh species occupied another type of habitat. Although both species of a pair shared the same type of habitat, the population peaks of each species of a pair presented a temporal shift. This suggests that niche separation is based both on ecological factors and interspecific competition. Factors favouring cockroach outbursts, and factors enhancing the coexistence of species are discussed.

Persistence time of apparent competition in a host-parasitoid microcosm is increased by metapopulation structure

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In order to quantify the role of spatial scale on the population dynamics of ecological assemblages, we use well-replicated, laboratory microcosm designed experiments. We have developed an experimental system containing an apparent competition interaction between two bruchid beetle hosts, Callosobruchus maculatus and C. chinensis, mediated by the shared parasitoid Anisopteromalus calandrae. The effects of metapopulation structures, of varying sizes, on the persistence of this apparent competition interaction are presented. The three species interaction persists longer in the spatially explicit environments where dispersal between patches is limited when compared with freely dispersing populations. We also show that the relative strengths of the ecological mechanisms of density dependence and parasitism that underlie this increase in persistence time alter as a result of the scale over which the interaction is observed changes.

Abundance, seasonality and biological aspects of Trypoxylon (Trypargilum) opacum (Hymenoptera; Sphecidae) at Parque Municipal Das Araucárias, Guarapuava (Pr), Brazil.

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Sphecidae wasps are mostly solitary and predators of a broad spectrum of insects and spiders. Their nests can be found in the ground, in hollow stems and pre-existing burrows, or made of mud and affixed to different types of substrates. The genus Trypoxylon is cosmopolitan however poorly represented in Australia and in the Pacific Islands. In this genus the greatest diversity is found especially in south and central America. The Trypoxylon biology has been studied in Brazil, but there are few information concerned to Trypoxylon (Trypargilum) opacum. Therefore, the purpose of this research was investigated the abundance, seasonality and some biological aspects of this species. It was collected 320 nests of T. opacum by trap-nests at Parque Municipal das Araucárias in Guarapuava (PR) Brazil, at 25o23´36´´S- 51o 27´19´´W. Nesting most frequently occurred during the hot season. Completed nests consisted of a linear series of brood cells ranging from 1 to 8, separated by mud partitions, usually followed by an empty vestibular cell and mud final-closure plug. Brood cells were provisioned usually with spiders of the family Araneidae. The number of females was always higher than males and they were randomly distributed. Natural enemies included Chrysididae, Ichneumonidae, Sarcophagidae, Bombyliidae and ants.

Physiologically structured host-parasitoid interactions: consolidating the physiological basis of increasingly complex models

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Physiologically structured host-parasitoid population dynamics models and behavioral models of host handling decisions – host feeding or oviposition - make different assumptions about nutrient acquisition and allocation to survival and fecundity in parasitoid females, leading to diverging predictions. While all these models are getting increasingly realistic, they now dangerously outgrow the experimental basis on which they were built. In this talk, we describe a model of lifetime dynamics of nutrients in a synovigenic, host feeding parasitoid. We followed a cohort of females over their entire life and recorded each oviposition and host-feeding events. Using micro-colorimetric methods, we then quantified the amounts of lipids, glycogen, sugars and proteins in emerging females, in eggs and at death. We also quantified the daily maintenance needs and the gains from host feeding for these four classes of nutrients. A daily nutrient budget, based on more than 20 experimentally measured parameters, combined with further experiments, enabled us to identify the elusive 'metabolic reserves' in this highly dimensional space, and to quantify host-feeding gains in terms of survival and fecundity. We end the talk with a list of approximations which we consider useful to pursue in modelling physiologically structured host-parasitoid interactions and others which we consider negligible.

Plant-plant interactions affect the behavior of herbivorous and predatory mites in a tritrophic context

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In response to herbivory, plants emit a specific blend of volatile compounds called herbivore-induced plant volatiles (HIPV). One of the ecological functions of HIPV is to attract the carnivorous natural enemies of herbivores. HIPV are also known to mediate plant-plant interactions. We studied these plant-plant interactions in a system consisting of lima bean plants (Phaseolus lunatus), herbivorous mites (Tetranychus urticae), and predatory mites (Phytoseiulus persimilis). Upon reception of HIPV released by T. urticae-infested plants, intact plants start emitting a blend of volatiles similar to HIPV. The release of volatiles by the receiver plants can be explained by a process involving adsorption of HIPV onto the plant's surface followed by its re-emission. Furthermore, upon herbivory, the HIPV-receiver plants emitted higher amounts of HIPV and attracted more predatory mites than unexposed plants. These data suggest that the reception of HIPV by lima bean plants can prime an indirect defense against herbivory. We also discuss the effect of plant-plant interactions on the behavior of herbivorous and predatory mites, and present data on their behavioral responses to exposed plants.

Interactions between habitat connectivity and population exchange of adult aquatic insects

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New Zealand's landscape has become highly fragmented over the last century with the clearance of large tracts of forest and conversion of land to agriculture. Understanding the permeability of river networks and landscapes to movement by aquatic organisms is important for planning the location of restoration activities aimed at enhancing aquatic biodiversity. To help understand relationships between landscape connectivity and population exchange of caddisfly and mayfly species, we investigated linkages between genetic distance and measures of terrestrial habitat quality and environmental distance (based on a range of climatic variables). Land cover data were divided into forested areas and non-forested areas, and connectivity metrics were calculated among pairs of sites. Several metrics showed relationships with genetic distance based on allozyme frequency for a hydropsychid caddisfly, with genetically similar populations generally occurring at sites with higher levels of connectedness than populations with "medium" and "high" genetic distance. Analyses of genetic distance among populations of a leptophlebiid mayfly species based on DNA markers suggested greater genetic distance among pairs of sites that were environmentally more distinct and where the intervening landscape was more patchy. We discuss the utility of this approach for interpreting connectivity/dispersal relationships over long (evolutionary) and short (contemporary) timescales.

Evolution of indirect plant mediated interactions in a community centered on Eurosta solidaginis (Diptera: Tephritidae)

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Indirect effects occur when the impacts of one species on another are influenced by one or more intermediate species. Modification of host plant traits by herbivores initiates indirect interactions between organisms that utilize the same host plant. We studied the evolution of indirect interactions mediated by plant traits in a community centered on galls induced by Eurosta solidaginis on goldenrod, Solidago altissima. Eurosta larval survival and the composition of the natural enemy community is influenced in a complex manner by gall morphology. Gall morphology shows strong geographic variation as the result of differing selection pressures. In part of Eurosta's range gall size is under stabilizing selection: small galls are attacked by the parasitoid Eurytoma gigantea, and birds attack large galls. In other parts of the range birds are absent and selection by parasitoids has led to the evolution of large galls. Large galls have in turn led to the reduced rates of Eurytoma parasitism and evolution of longer Eurytoma gigantea ovipositors. Altered gall size, and reduced Eurytoma parasitism has complex effects on other inquilines and parasitoids that attack Eurosta. Common garden experiments have shown that gall size is influenced by the interaction of plant genotype, insect genotype and the environment.

Spider effects on planthopper mortality, dispersal and spatial population dynamics

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Non-lethal effects of predators on prey populations scarcely have been considered in spatial ecological studies. Here, we report on the effects of spider predators on the mortality, dispersal and spatial population dynamics of the prairie planthopper, Prokelisia crocea. Based on survey data, the density of spiders declined significantly as host-plant patch size increased. Spider density also was negatively correlated with planthopper density per patch. Finally, planthopper extinction risk increased significantly with an increase in spider density. To evaluate whether spider effects on planthopper spatial dynamics were mediated by predation and/or spider-induced dispersal, we performed an experiment in which host-plant patches were either caged or left open and received different spider densities. For caged patches, planthopper recaptures declined only slightly with increasing spider density, suggesting that mortality effects of spiders on planthoppers were weak. In contrast, planthopper recaptures in open patches declined by 85% between the lowest and highest spider treatments. This decline was attributed to spider-induced emigration. We conclude that spiders have a greater impact on planthopper densities through induced emigration than consumption. The non-lethal effects of spiders may play a very important role in determining critical patch size, source-sink properties of patches, and the spatial distribution of planthoppers.

Plant-mediated competition in herbivorous insects: mechanisms underlying competitive superiority and consequences for population dynamics

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Feeding-induced plant resistance is a well-documented phenomenon for both sap-feeding and leaf chewing insects. Feeding-induced resistance via altered plant nutrition or allelochemistry provides a mechanistic basis for many cases of interspecific competition. An overwhelming number of cases of competition are asymmetric whereby one species is the superior competitor. Mechanisms underlying competitive superiority are diverse and complex. We review competitive interactions between phytophagous insects and determine the frequency of cases that are plant mediated. We also assess competitive superiority across several feeding guilds of insect herbivores and determine the specific plant-mediated mechanisms promoting such asymmetry. Drawing from our own studies we show that competitive superiority is linked to manipulation of plant nitrogen coupled with the ability to tolerate deteriorating plant quality via compensatory feeding. Moreover, competitive superiority is linked with reduced dispersal and the ability to tolerate diminished plant nitrogen. By contrast, colonizing species are poor competitors and meet nitrogen demands by dispersing to high-quality patches. Thus, plants as they interface with the life-history strategies of insect herbivores mediate competitive outcomes and dictate landscape-level differences in population dynamics. By integrating our work with published studies we determine the prevalence of plant-mediated competition and the constraints competitive superiority places on population dynamics.

Sex ratio in Melittobia parasitoid wasps: an exception to Hamilton's local mate competition theory

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Melittobia is a eulophid wasp explicitly identified by Hamilton as appearing to satisfy his ideal extreme inbreeding biofacies. Few quantitative data have been available to support this position, despite this genus being perhaps the most common parasite of a wide variety of solitary bees and wasps. In addition, the few studies published have not taken into account two important life history aspects: a mother plus up to 30 daughters combine their reproductive output on a shared host, and daughters develop in two morphological forms (morphs), long-winged dispersers and short-winged stay-at-home breeders. In a comparative study of five species of Melittobia, we placed groups of 1-5 colonizing long-winged females on both natural and factitious hosts, then tabulated offspring sex and form. Results did not support predictions for sex ratio adjustment with varying foundress number. Instead, sex ratio remained constant, with close to 98% females in the disperser morph. However, the sex ratio of the short-winged morph was much less strongly female biased, and increased as the number of females per host increased. Findings will be discussed in the context of current sex ratio models.

Predator avoidance and the ecology of multitrophic interactions

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Terrestrial ecologists have long investigated how predators locate their prey and the role of chemical information. This has resulted in extensive knowledge of the effects of chemical cues from arthropod prey and their food on the searching behaviour of predatory arthropods. In contrast, aquatic ecologists have historically concentrated on how prey use chemical cues from their predators to avoid being found and consumed by their predators. In the past few years ecological entomologists that are interested in terrestrial predator-prey interactions have started to pay attention to the effects of predator cues on prey behaviour and physiology. The data show that terrestrial arthropod prey also use chemical cues from their predators to avoid becoming a meal. Recent studies in our group have addressed the origin and specificity of the predator cues as well as the responses of arthropod prey to predator cues in relation to other environmental cues. The data show that we have long neglected the role of prey behaviour in the behavioural ecology of multitrophic interactions. In this paper I will address recent data as well as the major questions that lie ahead.

Are eastern North American monarch butterflies a homogenous group in terms of their migratory strategy?

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It was previously believed that the eastern North American monarch migrates only to discrete wintering colonies in Mexico. However, this monarch butterfly also migrates to Cuba. These two monarch migratory populations are not only different in their destinations, but in their strategies as well. Mexican migrants arrive at their over-wintering colonies in the Neovolcanic Belt at the end of October to early November. During their over-wintering, the butterflies amass on trees and are reproductively dormant, surviving on lipid reserves. In March, the butterflies mate and fly northwards. In contrast, Dockx's research suggests that Cuban migrant monarchs are not only different in their migratory destination, but in their migratory and reproductive behavior, mating strategy, and certain morphological traits. The Cuban migrants arrive in November to Cuba, but they do not over-winter, and most of them are not reproductively dormant. The majority of these Cuban migrants are reproductively active (90%). This implies that Cuban migrant monarchs could hybridize with the resident Cuban populations producing offspring in Cuba. This work reveals that the North American monarch butterfly migration phenomenon is more complex than originally thought, and that this influx of monarchs into the neotropics likely has a significant impact on the genetic diversity of monarchs in Cuba.

Genetic structure among dune lake populations in species with different dispersal mechanisms

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The perched dune lakes of Fraser Island, Australia represent naturally isolated habitats as they have never been connected to each other or to other freshwater systems. Inhabitants of these lakes must have had the ability to disperse in order to have colonised the lakes in the first place but the scale of ongoing dispersal is likely to be different for species with different dispersal mechanisms. This study assessed the population genetic structuring of two species, the dragonfly Orthetrum boumiera and the freshwater shrimp Caridina indistincta, using the mtDNA COI gene in 50 individuals per lake from five lakes separated by no more than 3.5km. The dragonfly O.boumiera, showed little population genetic structuring among lakes at this scale suggesting that aerial dispersal of the adults is maintaining gene flow among seemingly isolated populations. Conversely, populations of the freshwater shrimp C.indistincta showed a pattern of in-situ genetic radiation from the founding haplotype suggesting that inter-lake gene flow in this species is rare. This study suggests that even at very small spatial (<3.5km) and temporal (<500,000 years) scales, isolated populations may be evolving independently.

Using a top-predator to assess the effects of habitat degradation on invertebrate diversity

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In Western Europe breeding numbers of the Red-backed Shrike, an avian predator on large invertebrates and small vertebrates, decreased dramatically in the last century. We hypothesize that decreasing invertebrate availability led to lower prey availability and subsequently to declining Red-backed Shrike numbers. Availability of invertebrate species strongly differs in time due to phenology (seasonal effects), activity (weather and diurnal patterns) and peak abundance (between-year variation). Only a broad spectrum of potential prey species can maintain a continuously sufficient food supply. Our hypothesis was supported by a comparitive study between degraded (encroached) dunes, due to atmospheric deposition, in The Netherlands and an intact dune area, with low atmospheric deposition, in Denmark. In the dutch dunes only a few pairs remained, while in the danish dunes a vital population was present. Results showed that chick growth and fledging success was much lower in the Netherlands. Some groups of insect species were missing in the Dutch diet. Moreover, a shift to smaller prey species was found. We conclude that the effects of habitat degradation on invertebrate diversity is reflected in the reproductive success of the Red-backed Shrike. So a negative change in population status indicates lowered invertebrate diversity due to environmental stress factors.

Impact of the introduction of Digitonthophagus gazella (Coleoptera: Scarabaeidae) on the native fauna of coprophagous Scarabaeidae in Brazil

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Dung pads are a food source to many insects, mainly Coleoptera, Diptera and Isoptera, including several nematodes and flies associated with cattle, especially the horn fly, Haematobia irritans, one of the main pests of Brazilian cattle. Coprophagous Scarabaeidae (CS) while burrowing pads improve nutrient cycling, soil structure and forage growth for cattle, besides acting as efficient biological control agents of flies and nematodes of veterinary importance. Digitonthophagus gazella (DG) is an exotic CS to Brazil, and it was first registered at the UNESP Farm in Selvíria (Mato Grosso do Sul state) in 1993. Since 1989 CS are being weekly collected in a light trap in order to evaluate the impact of DG on the native fauna of CS. The diversity of species dropped sharply after DG invaded the area, which increased slightly three years later. For the majority of paracoprid species there was a reduction in the population density after DG was detected, while for some endocoprid species it increased. Some paracoprid species, considered common before DG colonized the area, are either not trapped anymore or in only small numbers. Ten years after DG invaded the Farm, it is still not clear if a new equilibrium was reached.

Testing the growth/differentiation balance hypothesis on willow

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The Growth/Differentiation Balance Hypothesis (GDBH) has been used as an experimental framework for examining environmental effects on secondary metabolism production. Yet, it has not been put to adequate test. We rigorously tested GDBH on two willow species, Salix eriocephala (rich in condensed tannins), and S. sericea (contains tannins as well as phenolic glycosides). In order to reveal potential temporal variation in response to environmental factors, we made four sequential harvests of cloned willow plants. We measured growth, rate of photosynthesis, and levels of carbon-based secondary metabolites on willows grown at a gradient of five soil-fertility levels (from no fertilizer to very high levels). There was remarkable correspondence between the two willow species in their physiological responses. Growth increased with increasing nutrient availability, as predicted by GDBH. Photosynthetic rate, as predicted, was less sensitive to nutrient availability than was growth. The predicted parabolic response of constitutive secondary metabolites across the soil fertility gradient received partial, but not complete, support. Perhaps our most important result was that the secondary metabolic responses were dynamic over time. The effects of nutrient availability on these willows seem to be dependent on the rate and degree to which plants acclimated to changes in resource availability.Density dependent processes in interactions between the parasitoid Microctonus hyperodae Loan and its host Listronotus bonariensis

Trait-mediated indirect interactions between herbivorous mammals and insects: claiming for an integrate view of the terrestrial communities

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Herbivorous mammals are ubiquitous and important plant consumers in most terrestrial communities. Mammalian herbivores, in addition to decrease plant density, can affect their resource in a variety of ways. In fact, due to the very plastic nature of plants, they can affect plants by modifying their life history, overall architecture, leaf morphology, the production of chemical compounds, and also by altering their spatial distribution (habitat selection). In an evolutionary time, mammalian herbivores can even act as selective pressure provoking long-term changes in phenotypic traits in plants. By sharing the resources with insects, these two very disparate organisms are often entangled in competitive interactions. My goal in this presentation is to assess the importance that the trait-mediated indirect interactions (TMII) play in the interaction between these two kind of herbivores, by reviewing the existing literature on both the diverse effects of mammals on plants and on competition between phytophagous insects and mammals, and by showing my own studies on mammal-insect-crucifer interactions. The main idea is that TMIIs are crucial for insects since in most cases 1) ungulates outcompete insects, and 2) the main short-term effect of ungulates is not a change in plant density but a modification of chemical, physiological, morphological and architectural traits of plants (induced resistance). I will also extend the consideration of TMIIs to those phytophagous insects like nectar-feeders or pollen-feeders that are mutualists of plants, specifically when both ungulates and insects select plants according to the same traits. In addition to the potential ecological consequences of TMIIs, I will argue that ungulates and insects can sometimes act as conflicting selective pressures on plants, since an evolutionary consequence of TMIIs has been little considered. All of these ideas suggest the necessity of considering together both phytophagous vertebrates and invertebrates in order to draw a more accurate picture of the dynamics of terrestrial communities.

Islands in a sea of sand and stone: genetic evidence on the role of flooding in the dispersal of mound spring wolf spiders in the deserts of South Australia

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Scattered around the margins of the Great Artesian Basin are numerous springs and seepages (referred to as mound springs in South Australia) forming a discontinuous arc of wetlands containing a diverse and specialised fauna. This fauna includes a number of endemic crustaceans and molluscs but is dominated by lycosid spiders. Of these Venatrix fontis is the dominant species in springs along the western edge of the Great Artesian Basin. This species is totally water adapted and cannot live away from free water. The vectors of dispersal available to these spiders are extremely limited, thus the springs can be considered analogous to islands. Exactly how these species move about has not been directly observed, however we present genetic evidence that shows that dispersal between springs is taking place and the importance of periodic flooding in the movement of these spiders from spring to spring. Allozyme electrophoresis and SSCP analysis of the ND1 region of the mitochondrial DNA show significant substructuring of spring group spider populations. Principle Components Analysis of Nei's genetic distance shows clustering of spring populations by to drainage catchment. Mantel tests support this and show that genetic distance correlates to distance along drainage channels rather than linear distances.

Parasitoid mark-release-recapture techniques: development of marking and trapping techniques for small parasitoids

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Small parasitoids of Bemisia argentifolii were marked with the purified protein, rabbit immunoglobulin G (IgG). When marked parasitoids were assayed using an anti-rabbit IgG ELISA, virtually all of the marked Eretmocerus sp. contained enough protein to be distinguished from unmarked parasitoids. A field mark-release-recapture study was conducted to examine the dispersal characteristics of Eretmocerus emiratus. Parasitoids were released on three separate dates into the center of a cotton field bordered by cantaloupe and okra. A total of 1388, 637, and 397 marked and unmarked wasps were captured in suction traps over the three trials, respectively. The majority of parasitoids were captured between 0600 and 0800 h. Our traps consistently contained more males which suggests gender-specific differences in the dispersal behavior of E. emiratus. The distribution of the marked parasitoids revealed two distinct patterns. First, almost all of the marked parasitoids recaptured in the cotton plot were in suction traps adjacent to the release site. Second, marked parasitoids were recaptured more frequently in distant traps located in the cantaloupe plot than in distant traps located in the cotton and okra plots, suggesting that the parasitoids were moving toward the cantaloupe plots.

Primary succession of Collembola near a receding glacier in Norway

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The Hardangerjøkulen glacier in central south Norway has been shrinking during the last 250 years. The Collembola fauna was studied in 25 plots along a gradient from newly exposed soil to 8000 years old soil. All soil samples were taken in Salix herbacea vegetation (at 0-3 cm depth) to standardize the habitat. Both the total density of Collembola and the number of species increased with increasing age of the soil. Collembola were very efficient colonizers. Among a total of 34 species, 14 were present in 30-45 year old soil, and 26 after 70 years. One species, Agrenia bidenticulata, occurred only at the very border of the glacier. The youngest soil was dominated by large, surface-active species (especially Isotoma olivacea), while smaller, deep-living species usually waited until an organic layer had been developed. After 50-70 years, I. olivacea was still abundant, but its dominance was shared by Folsomia brevicauda and F. quadrioculata. The structure of the Collembola community was astonishing similar in 70-250 year old soil compared to 8000 year old soil, with Tetracanthella brachyura as dominant species, followed by the two Folsomia species, I. olivacea, and Friesea truncata. Oribatid mites were much slower colonizers.

Effect of habitat loss and invasion by scotch broom (Cytisis scoparius) on rare butterflies of Garry oak meadows

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Within Garry oak (Quercus garryana) ecosystems in British Columbia there are a number of species that are rare and endangered, including several species of insects. Habitat loss and degradation are undoubtedly factors in the endangerment of many of these species, as only five percent of the original Canadian distribution of Garry Oak ecosystems remains in an undeveloped state. This project examines metapopulations of rare butterfly species to determine 1) the effect of loss and fragmentation of habitat on butterfly populations, and 2) the effect of degradation of habitat quality of Garry oak meadows due to the invasive weed, Scotch Broom. Identification of which characteristics of meadows (nectar resources, larval host plant abundance, meadow size / spatial location, and abundance of Scotch Broom) have the strongest effect on viability of populations of several different species of butterfly is the goal of this project. Two networks of meadows (~100 meadows each) were surveyed in Duncan and on Saltspring Island. GLM regression and ordination methods were used to define habitat quality. For example, there were significant positive relationships between butterfly populations and increasing meadow size, density of oak trees, and presence of certain flower species. These data were used to adjust for 'effective habitat area' and to parameterize a predictive metapopulation model.

Evolution of dispersal in butterfly metapopulations

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Recent studies of butterfly metapopulations have demonstrated evolutionary life-history changes in response to habitat change and climate change. An important component of selection relates to the spatial dynamics of the species, establishment of new local populations and persistence of old ones. In the Glanville fritillary butterfly (Melitaea cinxia), females in newly-established populations in a large metapopulation are significantly more dispersive than females in old populations. Furthermore, mobility increases with decreasing connectivity (increasing isolation) among females from new populations, whereas the opposite trend occurs among females in old populations. The observed patterns in female mobility were predicted by an evolutionary metapopulation model. We demonstrate with the model parameterised for the Glanville fritillary that increasing landscape heterogeneity increases variance in mobility among individuals in a metapopulation, supporting the general notion that complex landscape structure may maintain life-history variation.

Climate change and the phenology of European aphids

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Aphids, because of their short generation time and low developmental threshold temperatures, are an insect group destined to respond particularly strongly to environmental changes. Standardised, daily data on the abundance of flying aphids have been brought together from countries throughout Europe and spanning up to 40 years, through the EU Thematic Network `EXAMINE'. Relationships between aphid phenology and environmental data have been quantified using the residual maximum likelihood (REML) methodology. These relationships have been used with climate change scenario data to suggest likely changes in aphid phenology over the next 50 years. Strong relationships between aphid phenology and environmental variables have been found for many species, but they are notably weaker in species living all year on trees, suggesting differences according to guild in the factors influencing phenology. In general, the date of first record of species in suction traps is expected to advance, the rate of advance varying with location and species, but averaging 8 days over the next 50 years.

Microbial impacts on plant-herbivore interactions

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The role of indirect interactions in structuring communities is becoming increasingly recognised. Plant-mediated interactions between insects are now well established, but colonisation by fungi may also affect insect herbivores by modifying the suitability of a shared host plant. For example, mycorrhizal fungi have been shown to affect the palatability of plants to insect herbivores, whilst plant pathogens may alter both the nutrient and secondary compound content of plant tissue, with consequences for the behaviour and performance of insects. This presentation will review the most recent work in this area, before considering a specific example, namely the plant-mediated interactions between a fungal pathogen, Marssonina betulae (Dueteromycotina: Melanconiales) and an aphid Euceraphis betulae (Homoptera: Drepanosiphinae) on silver birch (Betula pendula). Aphids preferred to feed on birch leaves inoculated with the fungus and their population growth was positively correlated with levels of infection. Inoculated leaves contained higher levels of free amino acids which may explain the positive interaction between the fungus and the aphid. We conclude by highlighting priorities for future research in this topical aspect of multi-trophic interactions.

Going with the flow: plant vascular systems mediate indirect interactions between plants, insect herbivores and hemi-parasites

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Indirect interactions between organisms exploiting a shared host-plant are recognised as an important phenomenon in community ecology. Traditionally, research into these indirect effects has focused on interactions between different types of insect herbivores, but I will consider the interactions between sap-sucking insects and other organisms that exploit the plant vascular system, specifically hemi-parasitic plants. Parasitic plants have major impacts on plant community structure, but their effects on insect herbivores sharing the same host plant remain little studied and poorly understood. I examine interactions between a host, (grasses Poa annua and Poa pratensis), a phloem-feeding aphid (Sitobion avenae, Sternorrhyncha: Aphidae), a xylem-feeding plant-hopper (Neophilaenus lineatus, Auchenorrhynca: Cercopidae) and a hemi-parasite which obtains nutrients from the xylem by attaching to the host's roots (Rhinathus minor, Scrophulariacae). Both positive and negative interactions occur between the components of this system, mediated by alterations to the host vascular system and resource removal. Aphids improved the performance of the parasite because they led to an increase in root biomass and hence more opportunities for the parasite to connect to the host. In contrast, the parasite had a negative effect on the performance of both insects. This work demonstrates that plants mediate interactions between taxonomically diverse organisms.

Interpatch movement and edge effects in heterogeneous landscapes: the role of behavioral responses to matrix composition

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To date, there has been little empirical exploration of the mechanistic basis of matrix effects on either interpatch movement or the generation of edge effects in animal density. In an earlier study (Haynes and Cronin 2003), we found that the planthopper Prokelisia crocea exhibits higher rates of movement among host-plant patches embedded within a non-host grass (brome) than a mudflat matrix. Additionally, census data indicated that the planthopper exhibits edge effects (i.e., aggregates near the patch perimeter) in mudflat-bordered patches, but not in patches bordered by non-host grasses. Here, we investigate the underlying mechanisms of these matrix effects using individual movement trials. Whereas movement through mudflat was highly linear, movement was much more tortuous through brome. Within patches, brome edges were 3 times more permeable to emigration than mudflat edges. We suggest that the effect of matrix composition on the planthopper's interpatch movement rate is driven largely by differences in movement tortuosity within the matrix. Tortuous movement through brome likely increases the planthopper's rate of encounter with spatially aggregated host-plant patches. Furthermore, the effect of the matrix on planthopper edge effects can be attributed to the differences in edge permeability between matrix types.

Interactions between complementary sex determination and Wolbachia in Braconid parasitoids

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We discovered Wolbachia endosymbionts in field and long-standing laboratory populations of the parasitoid Habrobracon hebetor. Since this species is known to exhibit complementary sex determination (CSD), we tested the hypothesis that Wolbachia enables CSD by comparing diploid male production in Wolbachia-infected and experimentally-cured lines of H. hebetor. Diploid males were produced in both lines, suggesting that the Wolbachia does not enable CSD. Instead, standard curing/mating studies showed that the Wolbachia causes cytoplasmic incompatibility (CI) in H. hebetor. We developed mathematical models to investigate the effect of CSD on fixation of CI Wolbachia under various assumptions regarding CSD. The simulations showed that CSD has very little effect on the generations to fixation if diploid males die (as in H. hebetor), but that the time to fixation is increased if diploid males survive. Other parameters explored in this model included whether or not surviving diploid males are accepted as mates, whether eggs 'fertilized' by diploid sperm die or develop as haploid males, and whether uninfected eggs that are fertilized by Wolbachia-infected haploid sperm die or develop as haploid males. We also investigated the effect of mating structure (which is itself affected by CSD) on fixation dynamics of CI Wolbachia. We used this model to generate predictions regarding the distribution of C.I. Wolbachia among parasitoids with various sex-determining systems and mating structures. We evaluated these predictions with a survey of Wolbachia across parasitoids with known sex determination and mating structure. For this analysis, we used both known cases of known presence or absence of Wolbachia from the literature, and our own data on Wolbachia presence or absence from selected species.

To what extent do aphids act as extrafloral nectaries?

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Recent research has revealed that plants bearing extrafloral nectaries can respond to herbivory by increasing the output of extrafloral nectar or growing new extrafloral nectaries, often only on the part of the plant affected by herbivory. It is also known that phloem-feeding homopterans can be beneficial to their host plants if herbivory pressure is high and the homopterans are tended by ants that remove other herbivores. However, it remains unknown whether plants can manipulate the homopterans' honeydew output in response to damage by other herbivores so as to become more attractive to ants. In other words, can some plants use the aphids' honeydew output as an inducible defense in the same way as other plants use extrafloral nectaries? Here, we present laboratory results addressing this question using the Japanese willow Salix gilgiana and its ant-tended aphids (Homoptera: Aphididae).

Diseased interactions: how “hidden players” like pathogens mediate interactions between plants and herbivorous insects

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Microorganisms are well-known to affect interactions between plants and herbivorous insects. Infection of plants by phytopathogens may have beneficial, detrimental, or no effects on herbivores. Vice versa, herbivory may positively, negatively, or not affect the plant´s susceptibility to plant pathogens. A plethora of studies is available analysing the effects of tripartite interactions between plants, herbivores, and phytopathogens (1). However, little is known on the mechanisms. A study will be presented which shows that fungal infection of Chinese cabbage induces an increase of peroxidase activity which may be considered to cause negative effects on the mustard leaf beetle Phaedon cochleariae. Other fungus-induced changes of the cabbage´s primary and secondary metabolism obviously did not affect the leaf beetle (2). Furthermore, while numerous studies analysed interactions between plants, phytopathogens and herbivores, a fourth player has been neglected so far: the entomopathogens. Here we provide evidence that phytopathogenic infection of a plant enhances the susceptibility of herbivore larvae to an entomopathogen (3).Impacts of climate change on species’ range margins

Can mite-fungal mutualisms affect the population dynamics of bark beetles?

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Phoretic mites and fungi are common associates on the exoskeleton of beetles. A bluestain fungus, Ophiostoma minus, commonly carried by Tarsonemus mites, is hypothesized to out-compete fungi associated with the southern pine beetle, Dendroctonus frontalis. Here, we test whether antagonistic interactions between beetle-mycangial fungi and O. minus affect broad patterns in beetle population growth. We hypothesize that mutualistic interactions between phoretic mites and O. minus influence bluestain abundance within beetle infestations, and determine whether the mite-fungal mutualism is part of an endogenous feedback loop. Field surveys suggest that beetle populations declined sharply when mite and O. minus abundances are high. Experiments support evidence of a strong interaction chain: from mites to O. minus to beetles. However, O. minus and mite abundances appear to function independent of beetle abundance and may be driven, in part, by exogenous variables such as climate. Our results suggest that mite-fungal interactions can influence broad scale patterns in beetle populations.

Elevated CO2 and insect herbivores

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Plants grown at elevated CO2 have reduced nitrogen concentrations, increased C:N ratios, and often, increased concentrations of carbon-based defensive compounds. The impact of this reduction in resource quality for the performance of herbivorous insects has been the subject of over 60 experimental studies. Variation in responses, both between and within insect species, has been found, but some generalities have emerged. The ability of particular species or feeding guilds to exhibit compensatory feeding in response to poorer food quality appears to be the key factor influencing their performance. Leaf chewers feeding on whole plants appear better able to compensate for poorer quality than do leaf miners. Phloem feeders show variable responses with some species appearing to be advantaged under elevated CO2. Investigation of the impacts of elevated CO2 on herbivorous insects is increasingly being conducted under more realistic environmental conditions and in conjunction with other factors such as temperature, nutrients and light. Large questions remain, however, as to the relative importance of changes in climate, CO2-mediated changes in plant quality and impacts of natural enemies for herbivore populations in the future.

Population regulation and key factors responsible for population fluctuations of sunn- pest (Eurygaster integriceps Put.) in Varamin cereal fields

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Sunn-pest is the most serious insect pest of wheat and barley in most areas of Iran. Life table studies were carried out in Varamin (a county in the vicinity of Tehran) during 1998-2000. Because of a considerable distance (about 30 km) between fields and resting sites (nearby mountains), sampling was done separately in two places. Key factor analysis was carried out both for whole stages and submortalities. Adults, which usually spend a period of ca 9 months in diapause, suffered an overcompensatory density-dependent mortality. This was the first key factor contributing around 73% of the total variance of mortality. Other important mortality factors were egg parasitism by Trissolcus vassilievi Mayr, adult parasitism by tachinids, and adult loss of fecundity due to environmental constraints. Among them T.vassilievi had an important effect on fluctuations of the adult numbers, even though it had a minor contribution on overall variance of K. The adult mortality resulted by scramble competition along with the parasitoids, regulated population of the pest at a level around 81.7 adults/m2.

Indirect effect of a lace bug and disturbance on a chrysomelid beetle via host willow quality

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In the riverside willow forest of the Kinokawa river (Wakayama, Japan), the population size of a willow leaf beetle, Plagiodera versicolora, decreased drastically in summer and recovered in autumn. This seasonal population dynamics synchronized with the leaf phenology of host willows. At this site, first leaf flush occurred in early spring and these leaves were maintained until summer in which second leaf flush occurred. This second flush was a compensative response for damages by floods, which occurred in summer or early autumn, and enhanced leaf quality due to the increase in young leaves. Because leaf age is generally related with leaf quality for herbivores, the intrinsic rate of natural increase of P. versicolora might decrease in summer and increase in autumn. In addition, a lace bug, Metasalis populi, increased drastically in mid summer and decreased in autumn. This severe leaf damages by feedings of M. populi in summer might accelerate the decrease in host leaf quality for P. versicolora. In fact, P. versicolora adults fed on leaves damaged by M. populi could not lay eggs. This evidence suggested that indirect effects of M. populi and disturbance on the seasonal population dynamics of P .versicolora via host willow quality.

Arthropod community of a temperate non-tidal sandy beach: multitrophic interactions between washed-up seaweeds and insects in the supralittoral zone

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Sandy beaches are usually inhabited by numerous insect species with varying degrees of dependence on and adaptation to marine environments. At Hel Great Beach (northern Poland, Baltic Sea) a total number of 16465 of macrofaunal arthropods belonging to 30 species or genera was collected using the litterbags, and 32082 individuals from 61 species or genera by the use of the Barber's traps during three-year study (1999/2001). Coleoptera were the most numerous group, making up 67% of the total of identified arthropod specimens. The macrofaunal composition varied considerably not only from one sampling approach and locality to another but more from season to season. Ephemeral resource units, such as wrack-bed communities, were generally characterised by a successional change of species with different groups associated with different qualitative stages of decomposition and ageing of the stranded seaweeds. The major arthropod groups could be subdivided into "early", "mid" and "late" in relation to their appearance in the wrack. The successional colonisation of wracks was thus strongly influenced by the species' physiological and trophic needs and their appearance or disappearance in the wracks was due to the progressive microclimatic changes of the deposits in relation to their varying position on the beach.

Spatial variation in outbreak densities of Yponomeuta evonymellus in Finland

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A challenging task for ecologists has been explaining the spatial and temporal variation in densities of natural populations. The small ermine moth, Yponomeuta evonymellus (Lepidoptera, Yponomeutidae), is a specialist insect herbivore feeding on bird cherry. We have found records of large-scale fluctuations in Y. evonymellus densities in Finland since 19th century. During the peak years, bird cherry trees become totally defoliated. However, outbreaks are observed in only certain populations and within populations, there is considerable variation in larval densities even within adjacent trees. We examined the factors contributing to spatial variation in population densities of Y. evonymellus during increase and peak phases of population cycle. We evaluated the potential role of host plant distribution and quality, habitat type (open/forest), parasitism and oviposition behaviour in determining the spatial structure of Y. evonymellus populations during high density years. Our results indicate strong effects of small scale variation in factors related to habitat quality. This aspect is highly relevant when considering e.g. the effects of forest fragmentation on forest pest populations.

Bark and ambrosia beetle (Scolytidae)-fungal interactions on broadleaf trees in Japan

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The European elm bark beetle, Scolytus multistriatus, and its fungal associates, Ophiostoma ulmi, have been well studied, as economic pest causing Dutch elm disease. However, less is known about interactions between other broadleaf-tree infesting bark beetles and fungi. Recently, mortality of Pittosporum tobira trees has occurred in Japan. Beetle species involved in the mortality is Scolytogenes birosimensis. We have initiated studies to examine aspects of the biology of this insect, and present recent data on fungi associated with S. birosimensis. We also review results of our past and current studies on some species of scolytid ambrosia beetles, Xylosandrus spp. and Scolytoplatypus spp., and discuss symbiotic systems and the effects of fungi on the beetles.

When are grass endophytes plant-defending mutualists?

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Endophytic fungi are considered to be mutualists because (1) the fungus subsists entirely on resources extracted from the host, and in grasses also asexual dispersion by growing into the host's seeds, (2) while the host receives benefits through increased resistance to herbivores, pathogens and drought and flooding stress, and enhanced competitive abilities. Using meta-analyses, I reveal that the reasons for the strong mutualistic stamp of endophytes are largely historical and system-based. Although ecological, evolutionary, mycological and agronomical studies involving fungal endophytes in grasses have proliferated within past decades, most of the conceptual framework for endophyte-plant symbiosis has been based upon endophyte-plant-herbivore studies of two, economically important, artificially selected and introduced agricultural grass cultivars (tall fescue and perennial rye grass). Our studies (as well as few others) with native grass- and tree-endophyte systems reveal, however, that (1) endophyte-plant interactions are much more complex than described in past literature, (2) they represent a diverse continuum from antagonistic to mutualistic interactions, and finally (3) because benefits from endophytic fungi do not come without associated costs, the defensive mutualism may be conditional on available resources, particularly nutrients in soil.

Metapopulation dynamics of interactions between cannibalistic predator and its prey

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Here I propose a model involving three features, typical for many insect predator-prey interactions: (i) Insect pests are characterized by periodic outbreaks, during which predators are not able to regulate their numbers efficiently. (ii) Dynamics of many insect species is characterised by periods of local existence in a patch (e.g., larval period in butterflies, aphid colonies) periodically interrupted by periods of migration, during which prey and predator individuals redistribute themselves between patches. (iii) Cannibalism is common among insect predators and is often dramatically reducing their numbers. Nevertheless, it is usually ignored in models of predator-prey interactions. Here I assume that the predator considers any individual it meets (either prey or weaker conspecific) to be an edible item. The model based on these assumptions consists of a continuous-time within-season component describing population dynamics within one-patch, and of a discrete-time between-seasons metapopulation component involving between-patch migration. I use the model to predict the efficiency of such predator in controlling its prey and to study stability of this system.

Symbiotic secrets: reciprocity and antagonism in bark beetle-fungal interactions

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Reciprocal effects are commonly found between bark beetles and their fungal associates, and a diverse array of interactions, from antagonism to mutualism, is found within the group. Bark beetles vary in life history, but all feed in nutritionally poor substrates, especially phloem and cambial tissues of living or recently killed trees. Associations with fungi are apparently universal within this group. However, little is known of the contributions of fungi to bark beetle success, and even less is known about how these fungi interact with one another. We review the current state of knowledge on bark beetle fungal interactions and present recent data on the involvement of water potential, melanin and cellulases in these complex symbioses.

Overwintering strategy and mating behavior of Orius species

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Both males and females overwinter in many insect species. In Orius species, however, only female adults overwinter successfully while male adults cannot. Mendia scotti has been reported the female-biased survivorship during overwintering period. M. scotti males give nutrition to females through copulation, resulted in a low survival rate of the male. We supposed that Orius females also obtained nutrition from males through copulation, but before overwintering. To confirm this hypothesis, two experiments were conducted on Japanese Orius species. First, overwintering survival of females that had copulated in autumn was compared with that of unmated females. Second, copulation duration was compared between Orius individuals reared under autumn?like condition (OAC) and under summer?like condition (OSC) in a laboratory. If males of Orius species nutritionally contribute females' overwintering survivorship, the survival rate of mated females will be higher than that of unmated females, and the copulation duration in autumn might be extended rather than that in summer. In results of the experiments, the survival rate was higher in mated females than unmated females. Copulation duration was longer in OAC pairs than OSC pairs. These suggest males' nutritional contribution to females' overwintering survivorship in Orius species in autumn.

The influence of human land use on a diverse insect community – dung beetles in a relic tropical rain forest and surrounding farmland in Kenya

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Kakamega Forest in western Kenya is the easternmost remnant of the Guinean-Congolian rainforest. Embedded in a highly populated agricultural region, the area has been exposed to various types of anthropogenic degradation (logging, farming, wiping out the large mammals, etc.). As expected from general ecological preferences, dung beetle assemblages show a lower abundance in forests than in grasslands. The less disturbed forest has the fewest dung beetles, but a higher species number and diversity compared to a secondary forest. Grassland in an area where farmers collect 80% of the dung of their cattle for building and manure purposes has a much poorer dung beetle fauna than grazed grassland on a small farm where dung is not removed by men and is thus always available for beetles. The roller guild, generally very common in the Afrotropics, was almost completely absent all year round. Seasonal patterns were less pronounced in the less disturbed forest than in the more open habitats. Human land use influences dung beetle assemblages significantly, but in various directions.

Beetle-associated fungi and methyl jasmonate elicit tree defences and interfere with host selection and colonization in the Eurasian spruce bark beetle Ips typographus

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Pioneer bark beetles select their host through a combination of visual and chemical stimuli that mediate landing, inspection and tunnelling. In an acceptable host beetle development proceeds with mating, gallery construction, oviposition, and brood development. Phytopathogenic fungi vectored by the beetles elicit defence responses that may interfere with both host selection and beetle development. Chemical elicitors such as methyl jasmonate appear to have similar effects, inducing activation of phenol-rich cells in the bark and formation of traumatic resin duct in the sapwood. Induced trees seem to be less attractive to colonizing bark beetles and are more resistant to later mass-attacks. Induced host responses may inhibit beetle tunnelling by altering the chemical state of the phloem, and affect beetle survival directly through production of allelochemicals. These responses open up new possibilities for tree protection and management of tree-killing bark beetles.

Conservation strategy of Cicindela chinensis flammifera in Korea

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The distributions of tiger beetles in Korea have been rapidly decreased due to the urbanization of the countryside in past 30 years. Family Cicindelidae in Korea is consisted of 2 genus 16 species. Among them, 2 species are protected by law. However, many other tiger beetle species are facing a danger of extinction in result of the changes in their natural habitats. Ecology of Cicindela chinensis flammifera is studied for past 5 years in order to protect from possible species extinction. A conservation program including artificial mass rearing technique is developed, enabling a life cycle completion time from 2 years to about 7 months. Artificial ovipositing chamber and a rearing kit are developed. Up to 80 eggs can be collected from a single female, and 70% of them can successfully become adults. Best habitat condition including soil conditions, temperature ranges and space requirements has been experimented to implement the conservation program. A life cycle model is developed for the artificial mass rearing of other tiger beetle species.

The effects of climatic conditions on seasonal population fluctuation of date palm scale [Parlatoria blanchardi Targ (Hom: Cicadellidae)]

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Palatoria blanchardi Targ is a pest in date palm orchards of Iran. This investigation has been carried out from 1996 to 2000. The population was monitored monthly by leaf sampling. Sample unit was 3 leaves that taken in 3 strata in the palm canopy of ten trees. Climatic data (Temperature, Humidity and Rainfall) were obtained from Abadan meteorology station. Correlation analysis and sigmoid models were used to studying the climatic effects. The correlation of temperature (r=-0.58) and relative humidity (r=0.53) were Significant. The order of relative influences of climatic factors on seasonal population fluctuation was temperature> relative humidity> rainfall. The highest population fluctuation occurred in temperature between 16c and 24c and relative humidity between 42 and 63 percent. Date palm scale activities started at March and increased with temperature and humidity slowly. Seasonal monitoring showed three overlapping periods in field. First, second and third periods of activities started at March, April and December respectively. The longest period was the first period and the smallest was third. Scale nymphs were more susceptible to temperature and humidity than other developmental stages and their mortality increased at high temperature and low humidity. So, these stages get to aestivation at first periods of seasonal population fluctuation.

The use of water pan trap to estimate beetle abundance and vertical distribution in mosaic habitat in Kanazawa, Japan

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A continuous canopy sampling by using water pan trap was performed to observe beetle abundance, richness, color efficiency and vertical distribution in mosaic habitat in Kanazawa, Japan. A total of 845 individuals of which distributed in 47 families were collected from May to December 2000. Scolytidae was the most abundant (17.48 %), followed by Chrysomelidae (9.83 %) and then Cantharidae (9.14 %). Traps in bottom layer captured relatively high number of families and individuals, but the effect of vertical arrangement of traps did not differ significantly. Among the functional groups, herbivores and scavengers were shown to have a higher proportion in this layer. Some families were considered as layer specialists, but no clear stratum restriction was observed. Yellow trap demonstrated higher capture efficiency in beetle abundance than in blue trap. Effects of color were significant for herbivores ($P<0.01$) and predators ($P<0.05$), both were more abundant in yellow trap. Some families such as Attelabidae and Chrysomelidae showed a clear preference to yellow traps, but only Attelabidae was restricted to yellow traps.

Herbivore-induced responses in plant morphology and their impact on the third trophic level

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To free themselves of ectoparasites in nasty places, animals are known to display specific behaviour that promotes exposure of the parasites to their enemies. Plants tend to be regarded as more static organisms, but we report for the first time that they exhibit in essence quite similar behaviour. Tulip bulbs modify their internal structure in response to attack by a herbivorous arthropod (the rust mite Aceria tulipae). This herbivore is so tiny that it can move in between the scales of the bulbs. The resulting changes in distance between bulb scales are microscopic, yet sufficient to allow predatory arthropods (i.e. the phytoseiid mite Neoseiulus cucumeris) – three times larger in diameter than the herbivore – to enter the interior space of the bulb. The consequence of this plant behaviour is dramatic: with predators the inside of the bulb is cleared from herbivores, but without the bulb is eaten from within and dries out. The crucial changes in bulb morphology enabling predator access are controlled by ethylene, a plant hormone released upon herbivore attack. This was demonstrated in experiments on predator-prey dynamics in which the effect of ethylene was either promoted or blocked (using 1-methyl-cyclopropene).

Coexistence of competitors in a fragmented landscape: an experimental approach using a tropical insect and its parasitoids

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In the fugitive model for the coexistence of competitors, the superior competitor is a weaker disperser, and the inferior competitor is able to persist by using isolated or newly created patches. I studied a patchily distributed plant, a monophagous insect herbivore, and two specialised insect parasitoids that share this herbivore as their sole hosts in a tropical forest in Belize, Central America. I describe a combination of observational work (mapping the patch network and the spatial distribution of herbivores and parasitoids) and experiments (manipulating the degree of habitat fragmentation by destroying patches in different locations) to test the fugitive model as an explanation for the coexistence of these competing parasitoids, and to investigate how their abundance and incidence are affected by fragmentation. Spatial patterns of parasitism were consistent with the fugitive model. Furthermore, as predicted by the model of Nee & May (1994) and others, the superior competitor was disproportionately affected by habitat fragmentation.

Spatial dynamics of forest insect outbreaks

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Outbreaks of phytophagous forest insects are often spectacular, causing defoliation of millions of hectares of forests. A common characteristic in these outbreaks is the phenomenon of spatial synchrony among populations separated by as much as 1,000 km. By comparing patterns of synchrony among insect species exhibiting varying degrees of dispersal capabilities we are able to draw inferences about the relative role of dispersal vs. weather (the "Moran effect") as causes of spatial synchrony. Results suggest that weather is a more dominant cause of synchrony but the mechanisms by which weather function are often difficult to identify. There is also considerable evidence that the synchronizing effect of weather may be indirectly imposed via trophic interactions (e.g., predators or host populations). The 'new frontier' in the study of time-space data is the search for complex spatial dynamics (e.g., population waves). Application of novel statistical methods to historical larch budmoth data reveal has revealed the existence of outbreak waves in populations emanating from specific foci. These recurrent waves are associated with geographical trends in connectivity; simulations using a spatially explicit host parasitoid model suggest that these waves are the result of nonlinear reaction-diffusion processes interacting with variable habitat connectivity.

The response of Hemipteran assemblages to the restoration of Heather Moorland

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Moors dominated by Calluna vulgaris (heather) and other dwarf shrubs host distinct and geographically specialised invertebrate assemblages. The habitat is diminishing in the UK, largely due to high grazing levels. The resultant loss of dwarf shrubs produces a degraded, grass-dominated, plant community. Increasingly restoration management is being used to re-establish dwarf shrubs to such sites. We report the first study of the extent to which moorland vegetation restoration aids the reassembly of associated insect assemblages. Hemiptera were sampled by suction apparatus in grass dominated (degraded), restored and long established dwarf shrub (target) areas at each of eight restoration locations. Vegetation composition and soil characteristics were measured at each sampling point. Hemipteran assemblages differed between degraded and target sites. Species richness was significantly higher at the degraded sites whilst target sites hosted more specialised species. Samples from restored sites were, in all cases, more similar to their corresponding target than to samples from degraded sites. Plant community composition was the most significant predictor of hemipteran assemblages. Of measured soil characteristics, pH was the most significant predictor. This project demonstrates that restoration of dwarf shrub cover on degraded sites does aid the re-establishment of hemipteran assemblages typical of long established heather moors.

Can we predict insect community responses to climate change? Examples from grassland ecosystems

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It is increasingly apparent that individual species and, indeed, genotypes within species can respond differently to environmental change. However, to assist prediction there is a need to attempt generalisations, particularly with respect to population and community responses. Through investigation of the responses of insect herbivore communities on limestone grassland to the field manipulation of local climate, certain groups of species/traits appear to respond in a consistent fashion. The insect communities have been suction sampled regularly since 1993 on limestone grassland manipulated to reflect two climate change scenarios, warmer winters with either wetter or drier summers. There is a consistent gross response to summer drought while responses to winter warming are more species specific. Responses of the insect community dynamics and composition over the last twelve years have been tested against a simple trait based model of predicted general responses to climate change.

Responses of microarthropods to climate change on Marion Island: experimental evidence

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Our understanding of the direct, indirect and interactive effects of climate change parameters on invertebrates remains fragmentary. This is particularly true in the southern hemisphere. On Marion Island (Prince Edward Islands, sub-Antarctic), a significant reduction in mean annual rainfall and increase in temperature over the past half-century is evident. On this island we examined the effect of experimentally-induced drying, warming and shading on Collembola and Acari associated with a keystone plant species, Azorella selago (Apiaceae). Microarthropods were sampled from the centre of A. selago plants with an O'Conner split corer, and extracted from cores using a MacFadyen higher gradient extractor. In general, significantly more individuals were found in control compared to treatment plants, with no difference in species richness between treatments. Individual species responded differently to the treatments. These results suggest that microarthropod abundance on Marion Island will generally decline with ongoing climate change in the region, springtails will respond more strongly than mites, and at least one introduced springtail species will be favoured by these changes. Although no significant differences in species richness were recorded in this localized, short-term experiment, ongoing declines in species abundance across the island would inevitably result in species losses from the system.

Spatial population structure of Timarcha lugens: the importance of spatial autocorrelation

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Species inhabit patchy habitat can persist as metapopulation, define as a group of patchy populations connect by immigration. We studied the distribution of an endemic high mountain beetle living in a patchy environment, Timarcha lugens (Coleoptera, Chrysomelidae), in Sierra Nevada National Park (Southeast of Spain). For this purpose we study during five year 20 patches of the host plant. In addition, we analysed the importance of different variables to identify the factor to regulate this process, and the important of spatial autocorrelation. We used conventional analysis and non-conventional (Moran' I autocorrelation analysis and partial Mantel test to correct by the spatial autocorrelation) to determine the factor affecting beetle distribution. We found spatial autocorrelation in most explanatory and dependent variables and different results between conventional and non-conventional analyses. The main factors explaining T. lugens distribution were the position of the patches in relation to the river, isolation and plant density. Our result also shown that the distribution of T. lugens could be better explains by a mechanism of source-sink dynamic. In conclusion, spatial autocorrelation was important in this study, the use of specific spatial analyses being necessary to identify the real factors that contribute to regulate T. lugens populations.

Climate change and community-level turnover in British butterflies

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Butterfly richness patterns in Britain are well described by climatic variables and as a result the current changes in climate have led to many species extending their geographic ranges. Climate change is also thought to be leading to systematic changes in species richness and the structure of communities. This has not, however, assessed for British butterflies or for any other species assemblage. Here we investigate changes in the structure of butterfly assemblages (generalist vs. specialist) between two periods 1970-82 and 1995-99. We examined changes in species numbers and species turnover at three spatial resolutions across the whole Britain (50, 20 and 10 km grid cells). Though total species number has increased across Britain between the two study periods, the number of sedentary specialist butterflies has decline in some areas. Therefore, butterfly assemblages have became dominated for more mobile generalist species at least in some areas of the country. We discuss the results in relation to current climate warming and their implications for the future of global biodiversity.

Fitness consequences of predator-specific anti-predator behaviour in spider mites: Should I stay or should I go?

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Prey are expected to adopt predator-specific strategies when antipredator behaviour is effective against one predator but not against another. In this work, we quantify the fitness consequences of antipredator behaviour of the spider mite Tetranychus urticae, in order to predict and test its antipredator behaviour when exposed to different predation risks. The analysis of antipredator behaviour requires the quantification of costs and benefits of these two conflicting actions in terms of a common currency. Since spider mites have locally unstable populations, yet persist in a network of local populations connected by dispersal (i.e. a metapopulation structure), an adequate measure for fitness is the number of dispersing offspring per female produced during a local predator-prey interaction. Using a simple predator-prey we predicted the number of dispersing offspring produced by a female when faced to different predators. This model predicted prey should adopt predator-specific strategies to escape extinction. Experiments in which prey could display both strategies confirmed these predictions. Consequences for population dynamics are discussed.

Experimental evidence for apparent competition in a tropical forest food web

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Tropical insect communities are some of the most diverse on earth, but the processes that structure them and determine their diversity are largely unknown. Apparent competition is one of the major candidate processes. We experimentally removed two species of herbivore from a community of leaf-mining insects in a tropical forest in Belize. We predicted that other species which share natural enemies with the removed species would experience lower parasitism and have higher population densities in treatment compared with control sites. In both cases we found significantly lower parasitism, and in one case significantly higher abundance a year after the manipulation. This is the first demonstration of long-term apparent competition in any terrestrial arthropod community and suggests that apparent competition may be an important structuring force in tropical insect communities. Our results also have implications for the impact of human activities on tropical forest biodiversity.

Insects in a warmer world: ecophysiology of reproduction and overwintering of the southern green stink bug Nezara viridula in a recently colonized area in Japan

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From 1960s to 2000 N. viridula expanded its range northwards in Japan and reached Osaka (34.7 N; 135.5NE) following climate warming recorded in the region. The timing of diapause induction and its effect on life-history traits were studied under quasi-natural conditions in Osaka. Adult diapause induced in September–October ensured high winter survival. The timing of diapause induction strongly affected overwintering success and post-diapause performance: Females that emerged and entered diapause in October survived longer and produced more eggs than females that attained adulthood in September. Females from the early series reproduced until November, although progeny from the eggs laid in late September died during winter. Pre-winter reproduction of adults that emerged in mid-September or later was a result of the imperfect timing of diapause induction. It is an ineffective allocation of resources and considered the ecological cost of range expansion. To establish in the region, N. viridula will probably evolve a lengthening of the critical photoperiod of the diapause induction. This will allow to enter diapause earlier and, thus, avoid maladaptive late-autumn reproduction, but, perhaps, increase the duration and cost of diapause. The response of the species to climate warming will be compared with those reported in other species.

The role of dispersal in the population dynamics of locally unstable populations of prey and predators, using spider mites and predatory mites as a model system

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The population dynamics of the two-spotted spider mite (Tetranychus urticae) and its natural enemy, the phytoseiid predator Phytoseiulus persimilis, is highly unstable, leading to frequent extinctions of either species at the scale of individual plants. In spite of this, coexistence at a larger spatial scale may still be possible provided that local extinctions and colonizations occur asynchronously. To understand the role of local versus regional dynamics in maintaining population persistence, a series of small-scale greenhouse experiments were conducted and a generic model was developed to simulate within- and between plant dynamics. The model incorporates the functional, aggregative and numerical responses of the predators to the local density of prey, the negative feed-back between the prey's growth rate and the state of the host plant, and condition-dependent dispersal of mites among plants. The model shows good agreement with the experimental data and provides a good analytical tool to investigate strategies for biological control of fugitive pest species by means of specialist predators.

Parasitoids on Acronicta rumicis in South Korea

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The endoparasitoids of Acronicta rumicis were studied from May 2000 to October 2002 at Yong-Un dong area of Daejeon city in S. Korea. The number of species and parasitic rates were recorded. The number of endoparasitoids was 8 Species in 2 Orders 3 Families : Glyptapanteles liparidis and Microplitis sp. belonging to Order Hymenoptera, Family Braconidae. Diadegma sp. and Mesochorus semirufus belonging to Order Hymenoptera, Family Ichneumonidae. Compsilura concinnata, Euexorista sp., and Exorista sp. belonging to Order Diptera, Family Tachinidae. Among them, G. liparidis and C. concinnata are well known endoparasitoids of Lymantria dispar. Of the 322 field-collected larvae of A. rumicis, 74 showed parasitoids (22.98% parasitism). The collected field record showed relatively high parasitism in May (39.50±34.04%) and in October (50.05±2.70%). In addition, Parasitism by Hymenopteran and Dipteran parasitoids was about 85.61±7.30% and 12.22±4.42% respectively.

A computer program for estimation of fertility life table parameters using jackknife and bootstrap techniques

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Fertility tables are powerful tools for understanding of the population dynamics of a species. Intrinsic rate of natural increase (rm) is the most important estimator derived from fertility life table analysis and describes the maximal rate of increase that is possible for the species under the given physical and biotic environment. To compare fertility life table parameters of different groups, it is necessary to have information on the uncertainty degree associated with its estimates. The rm program has been provided for entomologists who want to have precise estimations of the five main fertility life table parameters; R0, rm, T, DT, , and precision associated with them using both bootstrap and jackknife techniques. This program is the first specific program that allows calculations using bootstrap technique. The Bootstrap replicates and Jackknife pseudovalues are stored into an Excel file for ease of comparing of different groups using statistical tests. The rm program is prepared by using visual basic programming language and can be run under all windows operating systems.

About bisexual population dynamics

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Let be the moments of appearance of new generations and let's assume that on the time intervals there are the monotonous decreasing of individuals:

About periodic impacts onto isolated population dynamics

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In model of insect population dynamics it's assumed there exist moments which correspond to "moments of harvesting":

Indirect interaction webs of insect herbivores: consequences of plant-mediated indirect effects

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Since effects of herbivorous insects on matured plants are almost nonlethal but subsequently induce changes in various plant traits, herbivore-induced indirect interactions are ubiquitous in plant-insect systems. Nevertheless, the subject has not been considered as important as direct effects. I will illustrate how herbivorous insects are connected each other through plant-mediated indirect effects on the willow, Salix miyabeana. For example, the enhanced shoot growth as a compensatory response to spittlebug oviposition links spittlebugs and leafrolling caterpillars. The leaf shelters connect leafrolling caterpillars with aphids and ants, and the increased tending by ants links aphids and leaf beetles. Ant-mediated indirect interaction between aphids and leaf beetles is maintained by provision of leaf shelters. Hence, these plant-mediated indirect effects generate the interaction linkage of insect herbivores on the willow. The interaction linkage can be depicted using indirect interaction webs that include nontrophic indirect links. The indirect interaction web revealed more than three times as much as direct and indirect interactions involved in the willow-insect system. Consequently, the structure of indirect interaction webs on plants may be greatly different from that of the traditional plant-based food webs, and will provide a more realistic understanding of interaction networks in insect communities.

Role of aphid bacterial symbionts in resistance to parasitism

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Molecular diagnostic tools have recently shown that bacterial symbionts of arthropods are extremely common, yet the effects of these symbionts on their hosts are largely unknown. In addition to the well-characterized obligate primary symbiont, Buchnera aphidicola, which supplies nutrients lacking in the plant sap diet, the pea aphid, Acyrthosiphon pisum, harbours diverse facultative or 'secondary' symbionts (SS). Pea aphids may harbour combinations of at least five vertically transmitted SS, including three independent lineages in the ?-proteobacteria designated as the R-, T- and U –types, a Rickettsia, and a Spiroplasma. Two of the three types of symbionts tested (R- and T-type) confer resistance to the braconid parasitoid, Aphidius ervi, by causing mortality of developing wasp larvae. The consequences of infection with both resistance conferring symbionts (R and T-types) on the aphid host and developing parasitoid will be discussed. These findings will be related to the distribution of SS in natural aphid populations. This interaction between a symbiotic bacterium and a host natural enemy provides a mechanism for the spread and persistence of these bacteria, and may be an important component of the widespread variation in susceptibility of pea aphid clones to parasitoid attack.

Genetic diversity and oviposition host preference of Bemisia tabaci (Gennadius) (Homoptera; Aleyrodidae) associated with four crops in Ghana

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The grave importance of Bemisia tabaci as a crop pest worldwide is mainly due to its wide host range, heterogeneous damage and intraspecific variation. Over 20 biotypes have been reported with varying host preference and vectorial capacity. In West Africa, two biotypes of this pest have been described by molecular and ecological studies. However, greater understanding of host-pest population interaction is necessary for its sustainable control. This study investigated the host acceptance of cassava and okra biotypes of B. tabaci using landing, oviposition and development preference on eight crops. The cassava biotype significantly preferred cassava for landing and oviposition ($p<0.001$), but did not oviposit on okra. The okra biotype significantly preferred okra ($p<0.026$), oviposited and developed on all other hosts except cassava ($p =0.001$), while the cassava biotype did not develop on okra, cabbage, and pepper. Mortality was highest in the first-instar nymph stage while development time was longer on marginal hosts than the preferred hosts ($p<0.05$). Thus, the observed host spectrum of each biotype is wider than earlier reported by host transfer experiments and molecular genetic surveys. Cowpea, garden egg and tomato are additional shared hosts whose role as reservoir hosts and biotype interbreeding grounds should be investigated further.

Significance of microhabitat on metapopulation structure in spider mites

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Restricted migration and habitat fragmentation promote genetic differentiation between populations. Spider mites exhibit two major dispersal behaviours, crawling and aerial drift. Tetranychus urticae infests many herbaceous and woody plant families, being considered that they disperse between adjacent plants mostly by crawling. Panonychus citri is another polyphagous species, yet most of its hosts are woody plants. Therefore, P. citri faces more risks by dispersion between host plants than T. urticae. If so, population structures of P. citri and T. urticae may be maintained in different ways due to different dispersal system. We analysed gene flow of P. citri in Japanese citrus groves from a broad geographic level to the patch level using molecular markers (esterase, a microsatellite and COI) and acaricide resistance genes. At the broad geographic level, gene diversity decreased with decreases in area size and distance between populations. Within a locality, on the other hand, genetic divergence of mites between citrus trees within groves was higher than that between the groves. From these results, we concluded that P. citri frequently migrates between groves within localities but not across wider areas, and within a locality they migrate between groves by aerial drift rather than moving to adjacent trees.

Parasitoid impact on aphid populations in UK cereal crops and attempts at manipulation

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Parasitoids are a key component of the natural enemy guild that frequently prevents cereal aphids from reaching damage thresholds in the U.K. To be effective, they must be present in cereal crops early enough in the season to coincide with aphid colonisation. This early spatio-temporal synchrony between aphids and parasitoids in the crop can be disrupted by climatic factors and by crop husbandry procedures. Field data will be presented to demonstrate the effects on aphid population dynamics of disruption of parasitoid activity during the early stages of crop invasion. The results of attempts to increase parasitoid impact by manipulation of field margin habitats and the use of aphid pheromones to manipulate parasitoid spatio-temporal distribution will be described. Changes in the sex ratio of adult parasitoids in the crop, from female biased early in the season to male biased late in the season reflect dispersal patterns between crop and semi-natural habitats in response to host availability. Preliminary data indicate that the deployment of aphid pheromones can positively influence the spatial distribution of adult parasitoids, encouraging early movement into the crop from field boundaries.

Historical perspective on multitrophic interactions

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Community views of nature have prevailed since well before Darwin's time, which have emphasized interacting species at multitrophic levels. Sophistication in understanding has increased from blatant and direct effects, such as predation and competition, to the many more subtle, but critical, indirect effects emphasized in multitropic-level-interactions ecology today. Enabled by advances in phytochemical analysis, the field advanced into the roles of constitutive plant chemicals, and plant odors and exudates in the ecology of higher trophic levels based on plants. Indeed, body odors (BOs) of all kinds have proven to be significant, whether from plants or insects, or waste products, or damaged cells, such that the ecology of BO is a burgeoning field. However, the science of multitrophic level interactions has remained largely idiosyncratic, while the potential for synthesis and empirically-based theory has been underexploited. Some examples of areas of future synthesis worth exploring on multitrophic interactions include: 1. the geographic mosaic of coevolution; 2. spatial variation in interactions; 3. long-term studies; 4. trait-mediated indirect effects; 5. the genetic basis of complex interactions; and 5. macroevolutionary perspectives employing phylogenetic comparisons among related species of plants, insect herbivores or natural enemies.

You are what you eat? Investigating the link between plant nutrient status and the performance of insect herbivores

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Predicting the consequences of global climate change may involve direct effects on insects but is likely to contain a strong component of altered diet. Therefore the effect of climate change on herbivorous insects cannot be determined without understanding the effect on the plant. Components of climate change including water and nutrient availability impact on plant quality, in particular the phloem. Aphids are herbivores that feed almost exclusively in the phloem. We are interested in the way in which phloem composition affects their growth and reproduction. Our strategy has been to modify sieve composition environmentally by altering water (drought) and nutrient input to the plant. By exploiting molecular genetics in the model plant Arabidopsis we are also able to modify sieve element composition using knockout mutations. Phloem sap is extracted using stylectyomy. Aphid performance (Rm) and behaviour (EPG, honeydew clocks) can then be measured and correlated with changes in diet. This work reveals that simple relationships between sieve element composition (diet) and aphid performance are modified by complex behavioural and metabolic manipulation of dietary flux by the aphid itself. These data inform models attempting to predict the effect of climate change and suggest that current prediction models may lack necessary complexity.

Restructuring of an exotic fungus-fly mutualism after establishment

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Botanophila flies and the endophytic fungus Epichloë spp. that infect Pooidae grasses have a mutualistic interaction. During oviposition flies transfer spermatia between Epichloë mating types for cross fertilization. Flies lay eggs on fungal stromata and larvae feed on the fungus. Studies on Epichloë spp. infecting wild grasses indicate that, in the absence of the fly, perithecial development following fertilization is reduced considerably. We were interested in examining the fungus-fly relationship after E. typhina was inadvertently introduced into Oregon in the United States. We observed that perithecia were common in D. glomerata but there was variation in the presence of the fly. At 12 sites we observed 6 to 98 % infestation of stromata (n = 50 per site) with fly larvae. At the 13th site, no fly eggs or larvae were detected on stromata but near uniform orange perithecia were found. An exclusion study conducted in a cultivated D. glomerata field also indicated that fertilization occurred without the fly. These results suggest that Botanophila could play a role in E. typhina fertilization in Oregon but it is not the sole factor involved. Hence, while Botanophila benefits from its association with E. typhina, the benefit to the fungus is questionable.

Host defense and parasitoid reproductive strategies limit niche breadth in a host-parasitoid system

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Defensive behaviour of larval Chrysophtharta agricola and host-stage preferences of two tachinid flies that parasitise it (Paropsivora australis and Tachinidae sp.) were examined. Data collected was used in conjunction with the results from other studies we conducted to establish the extent that host defence, host developmental biology and parasitoid reproductive strategies are impacting on the niche breadth of the two tachinids. Our data shows that Tachinidae sp. takes a relatively long time to oviposit and prefers early instar hosts which have less vigorous defensive behaviour than the final fourth instar, which thrashes its abdomen with sufficient vigour to dislodge the fly and interrupt oviposition. P. australis however, prefers to oviposit on fourth instars but as it can deposit an egg very quickly it effectively avoids being dislodged during oviposition. We discuss these results in relation to developmental interactions between host and parasitoids, and suggest that intraspecific host choice of these two tachinid flies is a result of interactions between both the biology and defensive behaviour of the host, and the reproductive strategies of the parasitoids.

Compositional changes of functional feeding groups in mountainous streams in a temperate region

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The ecosystem attributes of mountainous stream were analyzed using functional feeding group (FFG) ratios in three lotic systems in Korea. As the elevation went down from 800m to 400m, aquatic insect community structures were also changed and reflected the different ecological characteristics of each stream section. Although total biomass of communities in 3 streams were distinctively different, the longitudinal changing patterns of FFG ratios demonstrated tendencies that might be generalized. The ratio filtering collectors to gathering collectors was less than 0.5, and its values at higher elevations (alt. 800m & 600m) were very lower than those at lower elevation (alt. 400m). The ratio indicated the substrate stability was higher at both upstream and downstream sites comparing with the ratio at mid-stream. The ratios indicating the trophic type were much lower than 0.75. These values decreased along with elevation changing from 800m to 400m. The proportion of predators at each elevation also decreased from upstream to downstream. The ratios shredders to total collectors indicated that normal shredder association linked to functioning riparian system occurred at mid-stream. This study provided essential information in building the hypothetical river continuum model and generalizing FFG functions for the mountainous streams.

Two genetically distinct lines co-exist in populations of the endoparasitoid Venturia canescens

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In a laboratory colony of the solitary endoparasitoid Venturia canescens (Hymenoptera: Ichneumonidae) two genetically distinct lines (named RP and RM) appear to coexist sympatrically. The two lines differ in a cluster of phenotypic characters, including ovarian morphology, parasitism behaviour and reproductive success. Controlled oviposition experiments showed that RM females produce fewer offspring under single egg parasitism but more under con-specific superparasitism, compared to RP wasps. Investigations of interlarval combat under in vitro conditions showed that the higher reproductive success of the RM line under conspecific superparasitism is due to a physiological difference between the newly hatched larvae of the two lines which results in an advantage to the RM larva independent of the order or time interval between ovipositions. Examination of field-derived wasps has found that wasps displaying the two phenotypic clusters occur in natural populations. A model, using an iterative approach with experimental data to predict the stable composition in a mixed population of wasps displaying RM and RP, suggests that the two lines will coexist sympatrically when overall rates of superparasitism are greater than 5 to 10%.

Possible involvement of microorganisms in the production of Tetranychus urticae-induced lima bean leaf volatiles

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Lima bean leaves infested by spider mites emit volatiles that attract carnivorous natural enemies of spider mites. The predator-attracting volatiles consist of (Z)-3-hexenyl acetate, terpenes and methyl salicilate (MeSA), and their qualitative and/or quantitative composition has been shown to affect the atractiveness. The author has found that acidic as well as basic PR-protein coding genes were expressed in lima bean leaves in response to the infestation by the spider mites Tetranychus urticae. Since the acidic PR genes are generally induced by microbial attacks, we inferred that microorganisms that were pertinent to the spider mites might play a role in the volatile production in the infested leaves. Thus the microbial population in the mites was reduced as much as possible by rearing them under the aseptic condition, and their capability of eliciting the volatile production was investigated. The amounts of some of the volatiles from the lima bean leaves infested by the "aseptic" mites was markedly reduced compared with that in the leaves infested by the normal mites, suggesting a contribution of the microorganisms to the volatile production.

Statistical analysis of movement data in a heterogeneous landscape and behaviour at patch edges

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We used movement paths of adult bog fritillary butterflies Proclossiana eunomia to study the consequences of behaviour at habitat patch boundaries on dispersal in two habitat networks differing in fragmentation and matrix quality. Butterflies systematically engaged in U-turns when they reached a boundary in the fragmented network while they crossed over boundaries in more than 40% of boundary encounters in the continuous one. Applying the Virtual Migration model (Hanski et al Ecology 2000) to capture-mark-recapture data collected in both networks, we showed (1) a lower dispersal rate and (2) a lower survival during dispersal in the fragmented network. This latter difference is likely to be the key biological process leading to behavioural avoidance of patch boundary crossings. Then we designed an individual-based simulation model to explore the relationship between patch area, boundary permeability and emigration rate. Predictions of the model fitted observed results of the effect of patch area on emigration rate according to fragmentation: butterflies are more likely to leave small patches than large ones in fragmented landscapes (where patch boundary permeability is low), while this relationship disappears in more continuous landscapes (where patch boundary permeability is high).

The influence of transmission modes, host wasp and host tree preference on the population structure of Amylostereum symbionts of siricid woodwasps

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Siricid woodwasps (Hymenoptera) live in an obligate symbiosis with species of the fungus Amylostereum (Basidiomycotina). Wasps lay their eggs in softwoods together with a small inoculum of Amylostereum that causes a white-rot, where the wasp larvae develop. While there are many species of Sirex, only A. areolatum, A. chailletii and A. laevigatum are known to have symbioses with these insects. This difference in species numbers is reflected in specificity patterns: while a single wasp species typically is associated with one species of symbiotic fungus, the fungal species can be shared between different wasp species. Wasp species carrying the same fungal species commonly display different host tree preference. For example, S. juvencus and S. noctilio overlap in regional occurrence and both live in associations with A. areolatum. However, the former species preferentially breeds in Picea, while the latter shows a preference for Pinus. Here we studied the population structure of the Amylostereum symbionts of different wasp species, using phylogenetic and population genetic markers. We specifically determined genetic differentiation of symbionts between host wasps, host trees and geographical areas and inferred symbiont transmission modes. Transmission of the fungus between generations is usually thought to be strictly vertical from mother to offspring. However, the fact that various wasp species share the same fungal symbiont implies that horizontal transmission does occur, at least on evolutionary time scales. The observation of oviposition of wasps in the vicinity of previous Siricid and Amylostereum infections also suggests the possibility of both horizontal and vertical transfer.

Host range and population dynamics of greenhouse whitefly, Trialeurodes vaporariorum (Westwood) in North-Western Indian Himalayas

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Greenhouse whitefly, Trialeurodes vaporariorum (Westwood) (Homoptera: Aleyrodidae) has become a major pest of ornamental plants and vegetable crops under protected (greenhouse) and open environment in the temperate regions of north-western Indian Himalayas. The host range investigated in the region revealed the presence of greenhouse whitefly on 44 hosts belonging to 20 plant families. T. vaporariorum remains active through out the year on most of the hosts, however in open environment it passes the winter months (November-February) on Lycopersicon esculentum, Fuchsia magellanica, Salvia officinalis and Erigeron canadensis. The adult activity monitored using yellow sticky traps revealed two peaks of trap catch, one during May, both under protected and open environment, and the other in second week of September (under protected environment) and second fortnight of November (under open environment). Seasonal dynamics studied on brinjal, Solanum melongena, showed the peak activity of greenhouse whitefly during mid-June and September under protected environment, and early June under open environment. In the absence of biotic mortality factors in the region, the abiotic factors of the environment alone exerted regulatory influences on pest abundance.

Weeds and wasps: the effect of bridal creeper on parasitic wasp assemblages

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The invasion of exotic plants can significantly change natural patterns of biodiversity. In most cases the effect of weed invasion is measured as a change in the abundance of a single trophic level, usually native flora. However, it is less known what impact environmental weeds have on native fauna such as terrestrial arthropods and their associated interactions and ecological functions. One major arthropod group are hymenopteran parasitoids. The evolution of parasitoid-host associations has resulted in specific relationships with both host taxa and habitat. This study investigated the effects of one of Australia's most serious environmental weeds, bridal creeper (Asparagus asparagoides), on parasitic wasp assemblages in remnant eucalypt woodland in South Australia. Results consider parasitic wasp abundance, diversity and functional groups based on host associations, and reveal that although bridal creeper invasion has a strong negative effect on native plant diversity it does not reduce wasp biodiversity and supports an overall different wasp assemblage than adjacent native habitat. Given that over 450 parasitic wasp morphospecies were recorded, this study also highlights the high levels of arthropod diversity that can occur in a remnant habitat and the conservation value of small reserves for terrestrial arthropods.

The effects of elevated CO2 on insect populations and communities

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First, we present the results of a detailed study of the affects of elevated CO2 on different guilds of insect herbivores feeding on scrub-oak vegetation in Florida. Responses to elevated CO2 are compared both within guilds, for leafminers (6 species), gall insects (3 species), sap suckers (2 species), and chewing insects, and between guilds. Second, responses to elevated CO2 will be compared between and within guilds, and between habitats, using data from different elevated CO2 experiments at a variety of sites. We use meta-analysis for most of these comparisons. Finally, the effects of elevated CO2 is examined not just on selected populations, but on whole communities to see if insect community diversity is changed in elevated CO2. This analysis is based on newly collected data (2003-2004) from Kennedy Space Center and examines the effects of elevated CO2 on the diversity of plants, their herbivores, the herbivore's natural enemies, and the soil/litter dwellers. Because this is probably the only long term experiment examining the effects of elevated CO2 on a natural community, the results provide a unique opportunity to examine the effects of global change, in this case elevated CO2, on whole communities, not just populations of selected species.

Evolution in a community context: selection from herbivores and pollinators on plant traits

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All species are part of multi-species communities and the traits they express are shaped by selective pressures generated by the web of direct and indirect interactions among community members, along with the effects of abiotic factors. Understanding the diverse and simultaneous pressures experienced by species within a community may provide insights not readily gathered from the study of pair-wise interactions between species. Moreover, since the strength and direction of selection is altered by stochastic, abiotic factors that vary from year to year, a simultaneous examination of multiple selective forces carried out over many seasons is required to assess evolution in a community context. Continued experimental field manipulations of putative selective agents are combined with observational demographic data and with greenhouse experiments in this long-term study of the diverse agents acting on flower color in Raphanus sativus. Flower color and foliar glucosinolate expression are correlated, and thus both pollinators and herbivores may act as selective agents on both traits. In addition, to date, the effects of these agents appear to act in opposition, and vary in both strength and direction among years. In understanding how communities influence component species, we appreciate the contributions of biodiversity not only to ecological processes like ecosystem function, but also to the evolutionary trajectories of species. These trajectories are often shaped by the compromises imposed by simultaneous, diverse selective pressures.

How variable are responses of insects in the same region to climate change?

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Global environmental change is non-linear and impacting on insect management and conservation. The challenge for researchers is how best to identify the key issues that should drive management decisions on pest and non-pest species. Management decisions are usually site-based, and there are tensions caused by the desire to vary the emphasis put on each species. The response of a species to climate change or invasions by alien species will depend on its physiological, genetic and ecological attributes, and these will vary for each species at the same location. In addition, options for adaptation may be restricted by such constraints as pesticide resistance and non-target effects of pesticides such as endocrine disruption. A number of these attributes are identified and their implications discussed.

Molecular and ecological evidence for small-scale isolation by distance in an endangered damselfly

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Coenagrion mercuriale (Charpentier) is one of Europe's most threatened damselflies and is listed in the European Habitats directive that requires member states to designate Special Areas of Conservation for its protection. We combined an intensive mark-release-recapture study with a genetic analysis of 14 microsatellite loci for C. mercuriale from the Itchen Valley in the UK as part of an effort to understand the dispersal characteristics of this protected species with respect to its landscape. MRR data indicate that adult damselflies are highly sedentary. This restricted dispersal leads to significant genetic differentiation throughout most of the Itchen Valley, except between areas of continuous habitat, even though the core populations are separated by less than 10 km; this leads to a strong pattern of isolation by distance. Individual damselflies are positively autocorrelated at short distance classes reflecting fine-scale genetic clustering. A genetic patch size of 1.5 km is consistent with the distribution of movement events recorded by direct observation. These data are further discussed with respect to possible barriers to dispersal within the study site and the ecology and conservation of C. mercuriale. To our knowledge, this is the first report of fine-scale genetic structuring in any zygopteran species.

Invertebrate bioindicators to assess sustainability in vineyards

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The influence of management practices on invertebrates of economic interest and biodiversity in general provides important information for the development of sustainable agriculture. Invertebrates were sampled at ground level and in the canopy using three methods: pitfall traps and two types of sticky trap, to determine the effects of variation in two common vineyard practices (type of ground cover and level of irrigation). Earthworms were sampled by hand sorting soil samples. Results indicate that invertebrate taxa at the order level respond to management treatments. Order level biodiversity was increased with straw as a ground cover but was not affected by decreased irrigation. Ground cover (particularly straw) encouraged relatively larger populations of generalist predators and parasitoids as well as soil conditioning organisms such as Collembola and earthworms. Reduction in irrigation had a more limited effect, impacting on earthworm numbers and some generalist predators. However data also highlight some of the problems with the use of invertebrates for biomonitoring. In particular seasonal differences indicate the need for longitudinal studies incorporating a number of seasons.

Multitrophic plant-insect interactions and biodiversity at different spatial scales

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The focus in ecology is changing from the traditional study of simple interaction types to direct and indirect interactions among populations of multiple trophic levels. Such consideration of the complexity found in nature includes the realization that landscape structure directly affects local interactions and community structure and the need to study populations at different spatial scales. In fragmented landscapes, the disruption of habitat continuity may be accompanied by a loss of species and a disruption of interaction strength between species, showing the functional significance of biodiversity. Local abundance of functional groups may depend on different spatial scales of the surrounding landscapes, and I will test whether populations of higher trophic levels are influenced by a smaller or larger diameter of the surrounding landscape circles, i.e. by a different spatial scale. In my talk, I will present results form experiments performed in our agroecology research group that include plant-herbivore-parasitoid as well as plant-pollinator interactions.

Plant-mediated interactions between below- and above-ground processes: decomposition, herbivory, parasitism and pollination

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Traditionally, below-ground and above-ground communities have been investigated separately even though both systems show high interdependence. The interactions between above- and belowground organisms have only recently become a major field of study. Many of these studies focus on the effects of decomposers and below-ground herbivores on plant growth and above-ground plant-insect interactions, but their effect on plant reproduction is hardly studied. Further, the relative importance and the combined effects of below-ground and above-ground herbivores on plant performance, plant-herbivore, herbivore-parasitoid and plant-pollinator interactions is almost unknown. For this presentation and book chapter we will shortly review plant-mediated interactions between below- and above-ground processes. We will show results of our factorial experiments with potted mustard where we manipulated root and leaf herbivores as well as decomposers to analyze effects on plant-growth changes, herbivory, parasitism, flower visitation, fruit set and overall plant fitness. In these experiments we focus on the real-world complexity of direct and indirect interactions that only recently have been acknowledged by ecologists. By integrating our work with that from other published studies we will show that the traditional study of simple communities and interactions need to take such multitrophic-level interactions into account.

On the coexistence of thelytokous and arrhenotokous populations of Venturia canescens

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Venturia canescens is an important model for laboratory studies on behaviour and physiology of parasitoids. However, little was known about its field ecology. Studies in Mediterranean France have revealed that sexually reproducing V.canescens occur together with thelytokous ones. Thelytoky is not caused by PI-Wolbachia, and extended genetic studies have shown that successful exchange of genes between sexual and thelytokous populations is extremely rare or absent. This raises the question how populations of the two reproductive modes can co-exist. We will show the results of field studies on distribution and laboratory studies on behaviour and life–history traits to show that important ecological differences exist between the two modes. A population model will be presented showing that the two reproductive modes can co-exist, even when the sexual form is competitively inferior, when dispersal of the sexuals is better, or when sexuals are better in host finding under natural conditions.

Butterfly metapopulation dynamics from the parasitoid's perspective

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The spatial population dynamics of parasitoids, including their persistence, is influenced by the dispersion and population dynamics of their hosts. Here I contrast the spatial scales at which host distribution and dynamics are important for two parasitoid species sharing a single host butterfly species, using data from field surveys, experiments and molecular markers. The host, Melitaea cinxia, lives as a classical metapopulation in the Åland islands of SW Finland. Local populations occupy about 400 of the 4000 habitable meadows in an area of 50 by 70 km. One parasitoid, Hyposoter horticola, is extremely dispersive, finding most hosts and parasitizing a practically uniform fraction of them at all spatial scales. This wasp has very little spatial population structure of it's own and does not appear to experience the metapopulation structure of the host. It contributes to host population dynamics only by equally reducing the size of each host population. In contrast, the parasitoid Cotesia melitaearum is sedentary and has strong spatial population structure. It can influence host metapopulation dynamics but is limited, by it's dispersal ability, to a few tightly clustered host populations. Presently C. melitaearum is at risk of regional extinction.

Spatially explicit host-parasitoid relationships: density dependence revisited

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Species typically have aggregated distributions. Aggregation in species abundance may also cause aggregation at higher trophic levels, e.g. parasitoids responding to their host. Methods of quantifying aggregation differ in the way physical position is incorporated, i.e. their degree of spatial explicitness. Traditionally, methods used to detect 'spatial density dependence' have not considered spatial references when quantifying aggregation in either the host or parasitoid attack rate. Host-parasitoid relationships warrant reconsideration with spatially-explicit approaches. First, the spatial non-independence of host-abundance counts dictate that their spatial position must be considered in statistical analysis. Second, because parasitoids specifically respond to the spatial pattern of aggregation of their host's abundance, a spatial, biologically relevant description of aggregation is important. Using a wild silk moth (Gonometa postica: Lasiocampidae) and its parasitoids, we test if spatial density dependence is detected using a spatially-explicit analysis and compare the results with traditional approaches. Spatially non-explicit methods found limited evidence for density dependent parasitism, while the spatially-explicit approach found several cases. Therefore, at the between-plant scale, the failure to consider spatial pattern in the host and parasitoid attack rate may cause spatial density dependence to go undetected, while its inclusion increases the power and biological relevance of analyses.

How microhabitat and watertype differ seasonally in their importance for aquatic invertebrates

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Why does a species occur at a certain place? Species occurrence can be linked to habitat characteristics operating on different spatial and temporal scales. For aquatic insects, the occurrence can depend for example on the presence of floating vegetation for egg deposition, nutrient availability or historical glaciation events. In addition, the suitability of a place can change seasonally. Therefore the importance of habitat characteristics can vary seasonally and between different life stages. To study the relative importance of habitat (waterquality, depth, size) and microhabitat (vegetation structure, food availability) macroinvertebrate assemblages in three permanent ponds differing in acidity, nutrient availability and vegetation were studied. All microhabitats present in the three ponds were sampled separately on four occasions (spring, summer, autumn, winter). Detritivore, herbivore and carnivore abundance differed seasonally for the different watertypes and microhabitats, indicating differences in resource quality and quantity. These changes either force species to migrate to suitable places or have their life cycle synchronised to suitable periods. Therefore knowledge on the habitat and microhabitat use of species provides insight in a species life history.

Plant defence: linking above- and belowground multitrophic interactions

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Plants form the major link between above- and belowground multitrophic interactions. Research on multitrophic interactions aims at identifying the forces that regulate populations of species and structure communities. The study of direct and indirect plant defence is central to multitrophic theory. Direct plant defence relates to bottom up forces while indirect defence, where plants enhance herbivore control by natural enemies, can be considered an integration of bottom up and top-down forces. Recent results show that plant defensive chemicals can be vertically transferred through the food web, affecting organisms several trophic levels higher. The majority of plant defence work has focussed on aboveground interactions, while plants are challenged to defend themselves against both above and below-ground attackers. As yet we know very little of the ecological and evolutionary consequences of plant defence in an above-belowground multitrophic setting. However, there is increasing evidence that underground herbivory can induce aboveground plant defence responses (e.g. the level and distribution of plant defensive chemicals and foliar extrafloral nectar production), thereby affecting the performance of aboveground herbivores and that of higher trophic levels. Including belowground interactions in future research on multitrophic level interactions may shed a more realistic light on the role of plant defence in community ecology.

The use of functional groups to investigate community response to environmental changes.

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Independent studies of single species neglect indirect environmental effects mediated by species interactions, and so often fail to produce significant ecological insights. To deal with interactions in multispecies communities approaches covering all trophic levels have to be applied. Complex community data can be condensed without substantial loss of important information by reducing species composition to functional-group composition. Assuming that such functional groups differ in their response to environmental changes, links between environmental patterns and ecosystem functions can be revealed. We used empirically defined functional groups of plants (based on life-form traits), invertebrate herbivores, predators and detritivores (based on life-form and resource acquisition traits) derived from long-term data of two grassland communities to asses the impact of climate variation and anthropogenic disturbance by a phosphate fertilizer factory. Multivariate statistical techniques were used (redundancy analysis, simple and partial Mantel tests) to extract ecologically significant information from patterns in functional groups. This analysis resulted in two important findings: (a) trophic levels differed in their sensitivity to climate variation and (b) disturbance reduced the amount of interactions (connectance) in grassland communities. Thus, functional groups can be a useful generalization tool to study environmental effects on ecological communities. Optimum choice of traits to allocate species to functional groups by objective methods (e.g. statistical grouping techniques) might produce even clearer results.

Spruce budworm biotype reactions to forest and climate characteristics

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We review the population dynamics of eastern and western biotypes of the spruce budworm, Choristoneura fumiferana [Lepidoptera: Tortricidae]. These biotypes differ in the spatio-temporal patterns of their population dynamics as revealed by long-term defoliation histories. Examination of several population parameters revealed that western biotypes are more critically dependent on host synchrony than eastern types. Egg weights and survival to the feeding stage are critical parameters determining population success in the west. In the east, survival of large larvae and pupae determine generation trend. We relate these differences to two key (and interacting) environmental drivers: forest and climate characteristics. The forest characteristics include species composition, stand age distribution, and forest continuity over the landscape. Climate is important throughout the life cycle, but it is differences in the severity and consequent effects of late spring frosts that have had particularly dramatic effects on the insect species' population dynamics. In our concluding synthesis, we discuss the degree to which differences in the population dynamics of the two biotypes are rooted in genetics or environment. We also comment on the Pleistocene history of populations and discuss possible Neopleistocene changes to ranges of the insects in North America.

Invasive insects and climate change.

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Alien invasive species have been identified as a significant component of environmental change and intense international effort has been directed at their exclusion and control. Invasive insects can have major impacts on both natural and agricultural ecosystems as well as on health. However, the identity and impact of invasive species may alter under the influence of another component of environmental change, climate change. It is therefore imperative that prevention measures take these future interactions into account. Research has suggested that climate change may favour species with certain sets of traits. Therefore, we have attempted to identify broad generalities in the characteristics of invasive insect species found in several distinct case-study areas covering a range of climates. Information collected on the insects was subjected to a simple meta-analysis. We compare the results from the different regions to assess the affects of climate and also predict which characteristics may be favoured by climate change in the future. The possible implications this may have for prevention and management of alien insect species are outlined.

Using a mesocosm to test pattern and process in a Drosophilidae assemblage

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Spatial analysis is a tool whereby mechanistic hypotheses may be generated regarding spatial pattern. We investigated the spatial patterns of an assemblage of vinegar flies (Diptera: Drosophilidae) inhabiting a mesocosm to determine the mechanisms generating pattern. The mesocosm comprised a heterogeneous environmental surface differing in microclimatic temperature. Multiple spatial methods were used to quantify pattern and generate mechanistic hypotheses. Although only 21-36 % of the variability in the species and assemblage variables was spatially structured, the detected periodic spatial pattern matched the predicted response of the flies to the environmental surface. Structuring resulted from a combination of colonisation preference by the flies for shaded plots and increased mortality in sun, compared to shade, plots. Autocorrelation analysis also detected structure resulting from oviposition by flies across adjacent fruit and this was strongest across two fruit neighbours. An additional hypothesis relating to within-fruit oviposition patterns arose from the results of spatially explicit (SADIE) analyses. Therefore, although the spatial pattern was relatively weak in some cases and clearly generated by multiple mechanisms, spatial methods were able to accurately detect pattern and generate hypotheses concerning the mechanisms structuring this assemblage. A spatial approach can thus contribute significantly towards understanding the mechanistic basis of assemblage structure.

Carabid abundance in disturbed and rehabilitated riparian habitats in eastern Washington State, USA.

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Stream health and water quality are major regulatory concerns in the Pacific Northwestern United States. An integral component of rehabilitating disturbed waterways is the effective establishment of preferred vegetation in riparian buffers. We monitored three survey sites with varying degrees of ecological disturbance over a three year period in South Central Washington State. At each of the three survey sites, carabid beetle abundance was monitored in the riparian buffer corridor as well as in the adjacent orchard or vineyard. Our data shows that carabid beetle abundance is greater in highly disturbed plant communities comprised of exotic flowering plants. Plant communities comprised mostly of native vegetation with relatively low ecological disturbance contained fewer beneficial carabid beetles. Large stands of exotic flowering plants at ecologically disturbed sites hosted an abundance of pest insects. We conclude that carabid beetle abundance was greatest in these disturbed sites due to greater prey availability. Carabid beetle abundance was low in each of the adjacent orchards and vineyards studied. Cultural practices and pesticide inputs reduced the abundance beneficial carabid beetles in orchards and vineyards regardless of the ecological state of the adjacent riparian plant community.

Genetic structure of the endangered damselfly, Coenagrion mercuriale, in the UK.

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Coenagrion mercuriale (Charpentier) (Odonata: Zygoptera) is one of Europe's most threatened damselflies and is listed in the European Habitats directive that requires member states to designate Special Areas of Conservation for its protection. We have genotyped over forty of the major UK populations of C. mercuriale at 17 microsatellite loci as part of an effort to understand the genetic structure of this protected species with respect to its dispersal characteristics. There is significant genetic differentiation between most sites, even at small distance, that is consistent with MRR data that indicate this species is a poor disperser; the effect of this is a general pattern of isolation by distance. Gene flow (and presumably dispersal) is, however, enhanced by habitat connectivity whilst isolated populations, by contrast, can show marked skew in allele frequency, possible due to a founder event and/or bottleneck. These data are further discussed with respect to differences in genetic variation between populations and the conservation and management of this species.

Predicting the dynamics of new invasions based on fungal associates of exotic pine infesting bark beetles

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At least five species of pine infesting bark beetles (Coleoptera: Scolytidae) have been introduced into exotic plantations of the Southern Hemisphere. The best known of these are Orthotomicus erosus, Ips grandicollis, Hylurgus ligniperda, Hylastes ater and Hylastes angustatus. All but one of these species is native to Europe and I. grandicollis is native in North and Central America. Each of these bark beetles have been introduced into their new environments with a relatively large number of symbionts including a suite of fungi, most notably Ophiostoma spp. and their anamorphs. Studies on the taxonomy and population biology of the Ophiostoma spp. associated with introduced pine-infesting Scolytidae are revealing fascinating patterns relating to their introductions. These for example suggest that only a small portion of the mycoflora of the beetles in their native range, accompany them as exotics. Furthermore, overlapping niches of the insects in the exotic environment have led to changes relating to the fungi most commonly associated with them. These studies have shown clearly that further comparisons of the fungi associated with Scolytidae in their native and exotic environments are likely to be scientifically profitable.

Life system analysis of two coexisting Melitaenie butterflies in a patch network

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Two species of melitaeine butterflies coexisted in a patch network in Hebei, China. Based on analysis on butterfly mobility, genetic differentiation and population dynamics, Euphydras aurinia can be considered as a classical metapopulation while Melitae phoebe as a source-sink metapopulation. M. phoebe and E. aurinia, feed exclusively on their specific larval host plants Stemmacantha uniflora (Compositae) and Scabiosa tschiliensis (Dipsacaceae) respectively. The contrasting metapopulation structures and dynamics of the two butterfly species were determined by the interaction of the butterflies' behavior with the abundance and distribution of their specific host plants. The topographic features of the system (patch aspect and slope), sub-regional vegetation may explain the occurrence and distribution of the two butterfly species in the patch network. Parasitism does not play an important regulatory role in this system. Moderate grazing and patches embedded with small and discontinuous croplands promote patch occupancy and densities of larval groups. However, climatic variation (especially, drought in the early spring) is the main cause for the population fluctuations between years.

The conservation genetics of endangered mutualisms: increased genetic differentiation and reduced effective population sizes in specialist bees

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Complimentary sex determination in the haplodiploid Hymenoptera leads to the production of diploid males when diploid progeny are homozygous at the sex-determining locus. Diploid males are either inviable or effectively sterile, and their production increases the genetic load and drains hymenopteran populations of females. Fragmentation is expected to decrease gene flow within hymenopteran metapopulations resulting in reduced effective population sizes and increased levels of diploid male production. We explore the effects of diploid male production on the extinction of isolated hymenopteran populations, using bees as examples.

Average longevity of nymphal instars and hatching dates of grassland natural population of three species of grasshoppers

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Grasshoppers population was surveyed from June to December 2003 in grassland area in Durango Mexico, the most abundant species; Phoetaliotes nebrascensis (Thomas), Melanoplus lakinus (Scudder) and Boopedon nubilum (Say) were sorted by instar development and survey data. Exponential population models were obtained, taking dependent variable as grasshopper density and independent variables y and x as instar development and surveyed date ($P < 0.0001$). Midpoint in days of every nymphal instar was determined as the quotient of the integral of times x over the integral of , hatching periods were calculated as the integral limits of the exponential function over x with y equal to one, instar length ,of each specie was the slope the in linear regression of midpoints of presence of each instar and mean stages . The study showed a of 8.746, 13.142 and 5.861 days and hatching date on 16 July, 7 July and 27 July for P. nebrascensis, M. lakinus, and B. nubilum respectively. Mean values of life length were 15, 16, and 30 August for P. nebrascensis, B. nubilum and M. lakinus respectively, the bigger was 42 days for M. lakinus followed by 32 and 16 days for P. nebrascensis and B. nubilum.

Effects of fire on terrestrial invertebrates in buttongrass moorlands, Tasmania

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Buttongrass moorland is a significant landscape feature of western Tasmania. It is a unique ecosystem of world heritage value, poorly represented on mainland Australia and absent elsewhere. Buttongrass moorland is very inflammable, with a long history of firing by Aborigines. Since European settlement fire regimes have been substantially altered, and the role of fire in maintaining biodiversity in this ecosystem is largely unknown. The invertebrate fauna of buttongrass moorlands has not been well studied. Using pitfall trapping and sweep sampling, I found a terrestrial invertebrate community numerically dominated by Collembola, Diptera, Araneae, Acarina, Formicidae and Orthoptera. The most bio-diverse groups were Diptera (36 families, 290 species/morphospecies), Araneae (29, 214), Coeloptera (25, 98) and Acarina (35, 68). Using a before-after-control-impact experiment, I found that fire had little effect on species diversity, but a number of taxa showed significant numerical responses. Taxa that declined are those that inhabit the sparse emergent overstorey (eg Araneae: Thomisidae) or decaying vegetation (eg Amphpoda: Talitridae); these habitats recover slowly following fire. Taxa that increased include Orthoptera: Gryllidae, Coleoptera: Alticinae, Diptera: Chloropidae; these groups are probably responding to herbs and grasses that increased after fire. I will also present preliminary data on the effects of fire on population parameters (sex ratio, growth and reproductive success) for selected species.

So what are the impacts of environmental change on insect management and conservation?

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The main objective of this Symposium has been to examine the extent to which responses of insects to environmental changes are likely to be predictable. Speakers have attempted to consider and assess the possibility of approaching the issue by defining traits or combinations of traits that allow generalisation. To do this a range of comparative studies involving more than one species has been used. In this summary session we will attempt to answer a series of questions including: (i) if closely related species respond differently, why might this be?; (ii) if they respond similarly, can the generalisation be taken further? (iii) can we help each other, or are we on our own?; and (iv) what are the merits, drawbacks and complementarities of the range of methodologies that have been presented.

Impact of global warming and land-use change on insect management and conservation in temperate Asian area

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Different agroecosystems are inhabited by insect groups with different traits; resident/migrant, native/exotic, diapause/non diapause, uni/multi-voltine, and bisexual/parthenogenetic. The strategy of pest management contrasts sharply between greenhouse culture and rice. Greenhouse culture is a newly created habitat for temperate insects with short daylength and high temperature during winter. This may represent a model of an agroecosystem under global warming. Half of the pest species are exotic invasive pests originating in the tropics/subtropics, resulting in a great increase in insecticide applications. International trading has homogenized the fauna worldwide. Consequently there exist no targets to be conserved. Interception of alien pests by plant quarantine followed by IPM is needed. Paddy rice is not only the habitat of the rice arthropod community, but it is also an alternative habitat to the lost natural wetland for many endangered aquatic insects. Therefore, the management strategy should strike a balance between IPM and conservation, which is referred to as IBM (Integrated Biodiversity Management) (Kiritani 2000). In general, global warming may work in favor of natural enemies (except for spiders) by increasing the number of generations more than their host species. Biological control is expected to become a more important control tactic in the future.

Role of aphid bacterial symbionts in resistance to parasitism

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Molecular diagnostic tools have recently shown that bacterial symbionts of arthropods are extremely common, yet the effects of these symbionts on their hosts are largely unknown. In addition to the well-characterized obligate primary symbiont, Buchnera aphidicola, which supplies nutrients lacking in the plant sap diet, the pea aphid, Acyrthosiphon pisum, harbours diverse facultative or 'secondary' symbionts (SS). Pea aphids may harbour combinations of at least five vertically transmitted SS, including three independent lineages in the ?-proteobacteria designated as the R-, T- and U –types, a Rickettsia, and a Spiroplasma. Two of the three types of symbionts tested (R- and T-type) confer resistance to the braconid parasitoid, Aphidius ervi, by causing mortality of developing wasp larvae. The consequences of infection with both resistance conferring symbionts (R and T-types) on the aphid host and developing parasitoid will be discussed. These findings will be related to the distribution of SS in natural aphid populations. This interaction between a symbiotic bacterium and a host natural enemy provides a mechanism for the spread and persistence of these bacteria, and may be an important component of the widespread variation in susceptibility of pea aphid clones to parasitoid attack.

Grass Endophytes - Plant Mutualists?
K Saikkonen

Paper not available at time of production

Overwintering strategy and mating behaviour of Orius Species

T Kobayashi

Paper not available at time of production

Rapid change in settling behavior of the scale as an avoidance against parasitoids

T Matsumoto, T Itioka, T Nishida

Paper not available at time of production

Population regulation and key factors responsible for population fluctuations of sunn-pest (Eurygaster integriceps Put.) in Varamin cereal fields

Sh Iranipour, A Kharrazi Pakdel, Gh Radjabi

Paper not available at time of production

Genetic structure of the endangered damselfly, Coenagrion mercuriale, in the UK

P Watts

Paper not available at time of production

The use of water pan trap to estimate beetle abundance and verticle distribution in mosaic habitat in Kanazawa, Japan
AS Leksono, N Nakagoshi, K Nakamura, S Koji

Paper not available at time of production

The effect of salivary gland secretions on behavior of larval *Ephestia kuehniella* Zell (Lepidoptera: Phycitidae).

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The mediterranean flour moth, *Ephestia kuehniella* Zell. is one of the most cosmopolitan pest-species of insects and can be found in stored products and in the field. When the larvae meet they deposit a secret, which can regulate the oviposition behavior (Corbet, 1973a). In this communication it is shown that the extract of the larvae salivary glands secretions can have an effect on the larvae behaviour. In our investigation of the larvae behavior and the roles of taste and olfaction modalities in this were the Y-olfactometer and the method of repellent barrier used. First-, third- and fifth-instars larvae were used for experiments. The results show that the secretions of salivary glands has some active compounds, which can be felt by all instars larvae using taste and olfaction reception. The method of repellent barrier shows that in high concentrations the secretions of salivary glands acts as a repellent and causes escape behavior in all larvae instars. The olfaction experiments show behavior distinction between larvae instars. While in high concentrations of the secretions all instars show escape behavior, in low concentrations the first instar larvae has attractive response, stimulated by the secretion. As a result, these secretions can contribute to the regulation of larvae population density.

Volatiles from plant glandular trichomes of *Cordia curassavica* & their attraction to the herbivorous beetle *Cratosomus flavofasciatus*

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Glandular trichomes are epidermal plant appendages that accumulate a variety of low molecular weight chemicals, including monoterpenes and sesquiterpenes and have been recognized to serve in defense against herbivory. Behavioral, physiological and chemical evidences indicate that glandular trichomes on the New World species *Cordia curassavica* (Jacq.)(Boraginaceae) function in the attraction of the herbivorous beetle, *Cratosomus flavofasciatus* Guerin (Coleoptera: Curculionidae), especially at times of heavy plant transpiration. The emission of this plant volatiles may be an unavoidable byproduct of the transpiration process, is exploited by certain herbivores in locating their food source.

Host-orientation behaviors of *Lygus hesperus* (Heteroptera: Miridae) in response to visual and olfactory cues from plants and insect prey

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Lygus hesperus, the western tarnished plant bug, is found throughout the western United States and Canada, where it can cause serious damage to many agricultural crops. Interestingly, this insect can also serve as a facultative predator, and with this flexible feeding behavior it is possible that plant visual and volatile cues, as well as cues associated with potential prey may mediate host location and selection. A better understanding of the factors involved in host choice could facilitate the development of novel trapping techniques. To investigate the role of these various cues we tested nymphs and adults to headspace volatiles associated with host plants (alfalfa and/or cotton) singly or in combination with a visual plant cue (simulated by a green light-emitting diode) with or without potential prey (whiteflies or beet armyworm [BAW]). Laboratory bioassays were conducted with a Y-tube olfactometer and selected visual-volatile combinations were tested under field conditions. Headspace volatiles were identified by GC-MS analysis. Nymphs and female *L. hesperus* were most often attracted to plant volatiles; males were only attracted to volatile-visual cue combinations or to BAW-infested plants. Plant phenology, time of day, *Lygus* feeding and prey feeding habit influenced headspace chemical profiles.

Calling of sterile male Mediterranean fruit fly, *Ceratitis capitata*: effects of protein in the diet or exposure to ginger root oil.

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South Australia has recently incorporated the sterile insect technique (SIT) into its eradication programs for Mediterranean fruit fly, *Ceratitis capitata* (medfly) incursions. The possibility of improving sterile male Mediterranean fly performance through protein diets or exposure to ginger root oil (GRO) was investigated in laboratory observations on calling frequency of the Vienna7/Mix99 mediterranean fly strain. The experiments involved the use of cages containing either 10 males that were untreated controls, or 10 treated males. For the protein diet experiment, two trials were conducted with emerging flies being allowed to feed *ad lib* on either sugar agar or sugar agar and a dish of protein hydrolysate. Calling frequencies at two time periods on three consecutive days, analysed with repeated measures ANOVA, were not significantly different between protein-fed and control groups. For the GRO experiment, two trials were conducted to compare the calling frequency of untreated flies with that of flies exposed to GRO for 16 hours at age day 1-2. Repeated measures ANOVA indicated that the difference between treatments was significant in both trials with the GRO-treated males having a consistently higher frequency of calling. Studies to further assess the effect of GRO on behaviour are continuing.

Sand as a medium for transmission of vibratory signals of prey in antlions (Myrmeleontidae)

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Two European pit-building antlion species (*Euroleon nostras* and *Myrmeleon formicarius*) detect their prey by sensing the vibrations that prey generates during locomotory activity. For small arthropods walking on the sand a few centimetres away, the antlion larvae can determine direction of the prey's location. To observe signal transmission through the substrate we measured some of the physical properties of substrate vibrations in sand. The vibratory signals of four arthropod species (*Tenebrio molitor, Pyrrhocoris apterus, Formica* sp. and *Trachelipus rathkei*) were recorded in a distance of a few centimetres from an accelerometer. Frequency range of the signals varied from 0.2 to 4.5 kHz and acceleration values from 400 $\mu m/s^2$ to 1 mm/s^2. Pit-building antlion larvae are capable of discriminating between areas of sand differing in particle size, constructing pits in a fine sand and avoiding coarser sand. In sand fractions with different particle sizes (with sand grains up to 2.2 mm in diameter) we measured damping coefficients of the sand fractions for artificial pulses. Damping coefficient at frequencies from 100 to 300 Hz varied from 0.2 to 2.6 dB/cm and was inversely proportional to sand particle size and positively correlated with the frequency of the pulses.

Unpalatability of European checkerspots (Nymphalidae: Melitaeinae)

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Several Nearctic checkerspots butterflies, including the famous *Euphyyas editha*, are unpalatable to birds. In addition, checkerspots feed on plants with secondary metabolite with a protective function. However, palatability was never tested in Palaearctic Melitaeinae butterflies. We offered five species of checkerspots, *Euphyyas aurinia*, *Melitaea athalia*, *M. diamina*, *M. aurelia* and *M. didyma*, to Great tits (*Parus major*), using the satyrid *Aphantopus hyperanthus* as a control. The butterflies significantly differed in palatability (GLM, effect of species, $p < 0.01$). The difference was not between checkerspots and control, but within checkerspots. *M. aurinia* and *M. diamina* were eaten as often as control, *M. aurelia* and *M. didyma* were rejected. Wahlberg (Evolution 55: 522-537) showed that host plants of *aurinia* and *diamina* contain secoiridoids, while hosts of *aurelia* and *didyma* contain iridoids. Plant chemistry seems to be more important for palatability than phylogeny. However, a clear association between host chemistry and palatability is obscured by ambiguous results for *M. athalia* (iridoids). In any case, European checkerspots form a mimetic complex.

Protein expression in the reproductive organs of male German cockroach (*Blattella germanica*)

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Male accessory reproductive glands (ARGs) contain factors that are transferred with sperm during mating to the female. One such factor (sex peptide of Drosophila) inhibits female's receptivity to additional mating. Here, we analyzed ARGs protein content and pattern for males of the German cockroach (*Blattella germanica*). Lower protein mass were found in young males (<3day-day-old) when compared to older ones (3~10-day-old) by SDS-PAGE. This study shows the expression of genes in ARG was different with age, but the protein content of testis remained the same after adult emergence. In addition, western blot analysis and whole-mount immunohistochemistry were used to investigate sex peptide-like immunoreactivity in the reproductive organs of the male. Unexpectedly, the sex peptide-like substance was only found in the testis of *B. germanica*. The significance of this finding is currently under investigation.

Acoustic features of calling song in female mate choice in a field cricket, *Telleogryllus taiwanemma*

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Male field crickets, *Telleogryllus taiwanemma*, produce calling songs attract females. Variations in calling song phrase consisting of chirp sequences were recognized in Taiwan: phrases composed of single, double and seven chirps (Chow *et al.* 1990). However, Chow *et al.* did not show any relationship between the temporal structure in calling songs and behavioral significance, though variation in phrases may reflect, in part, male qualities. Using a Japanese cricket population having phrase variations in calling song, we tested whether females demonstrate preferences based on variations in calling song phrase using computer-synthesized songs. However, females did not discriminate between variations in calling song phrase consisting the same chirp rate. Females preferred higher calling song chirp rates; that is, the preference function shape was open ended. Preference function shape by females toward two other acoustic features of song chirp was also determined. Open-ended preference function was found in the pulse number of chirps, and unimodal preference was shown toward carry frequency, but which features correlate with male qualities remains unclear.

Pollinator responses to mimicry by sexually deceptive orchids

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Deceptive orchids mimic insect signals to attract pollinators without providing a reward. Orchid flowers may resemble food, oviposition sites, or female insects. Insects that respond, by attempting to mate with or feed from orchid flowers for example, collect and distribute orchid pollinia. Deceived insects may incur costs, e.g. time and energy wasted travelling to and interacting with orchids, and loss of sperm during pseudocopulation. Such costs may encourage the evolution of behavioural strategies that allow insects to avoid deceptive orchids. In sexually deceptive orchids, male pollinators are initially attracted by scents identical to female insect sex pheromones. Short-range cues eliciting pollinator contact with the flower may include visual cues such as colour and shape, and structural or tactile features that resemble female insect bodies. Investigation of the relative importance of chemical, visual, and structural cues, and the interaction between, long- and short-range cues should be rewarding. The Australian orchid genus *Cryptostylis* is an excellent study subject because all five species attract the same male wasp pollinator despite obvious differences in flower shape and colour. Here I introduce this intriguing study system and present some initial observations of pollinator behaviour and interactions between pollinator learning and the dimensions of orchid patches.

Characterization of the mandibular glands in different classes of adults in *Scaptotrigona postica* colonies

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Scaptotrigona postica, a Brazilian stingless bee, has paired mandibular glands, placed in each lateral head side, and constituted by a bifid sac, whose wall is covered by class III secretory cells. In *S. postica* workers, the mandibular gland produce a trail pheromone used by foragers to mark the pathway between the colony and the food source. The present abstract briefly describes the results of the histological and the morphometrical investigation of the mandibular glands in newly-emerged, nurse and forager workers, as well as in virgin, physogastric queens, and mature males of *S. postica*. The histology showed differences of the secretory cells related to the task of the bee in the colony. The morphometry showed that the mandibular glands have a single secretory cycle, in which the cell size is greater in the nurse workers, due to the secretion accumulation into the cells. Comparing the different bee classes studied, the physogastric queens have the greatest cells and nuclear areas, contrary to the males, which present the smallest cells. These observations suggest that the synthesis of the secretion starts from newly emerged and finishes in nurse workers. The queen is the class of individuals with more active glands. Although the function of the secretion was unknown in this caste, the present results clearly signalize their importance for it, since the queens presents more developed glands than the workers.

Comparative study of pupae cuticular hydrocarbon of flour beetles associated with their cannibalism and predation vulnerability (Coleopterae: Tenebrionidae).

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Cuticular hydrocarbon of pupae were extracted with n-hexane at room temperature and GC-MS (gas chromatograpy-mass spectometry) investigations were used to identify and compare pupae cuticular hydrocarbon composition of several species which have been previously quantified with n-eicosane as internal standard. The major components identified were n-C25; 3-Me-C25; n-C26; n-C27; 13-Me-C27; 3-Me-C27; n-C28; n-C29. However other molecules with low quantity were found in some species. Generally, qualitative and quantitative significant differences were observed in cuticular hydrocarbon found between the species. Cuticular hydrocarbon patterns seem to be correlated with cannibalism and predation vulnerability patterns. The most intriguing case was remarkably expressed by *T. brevicornis,* which pupae had the most of chemical compound found with the highest rate, but were unable to be cannibalised and predated by different perpetrators. This particularity rests on chemical signals, which could partly be due to variations in the proportion of certain cuticular hydrocarbons.

Studies on the dorsal abdominal glands & exuviate of local scentless bugs, *Leptocoris abdominalis* (Fabricius) & *Leptocoris augur*

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Two dorsal abdominal glands (median and posterior glands) are found in the nymphs of two local scentless plant bugs (Rhopalidae), whereas only the median glands are found in exuviate of nymphs. Both median and posterior glands are functional in nymphs, however, only posterior glands are functional in adults for the two species. Monoterpenes are the major compounds in posterior glands, but alkenals are the major compounds in median glands. Possible reasons for the sole presence of alkenals in exuviate will be discussed.

Body-wax sex pheromones & their functions in reproductive behavior of the black-winged fungus gnat, *Bradysia paupra* (Diptera: Sciaridae).

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A typical male mating behavior sequence of *Bradysia paupra* is: 1) intermittent wing fanning, 2) walking in a zigzag approach while continuing to fan wings, 3) abdomen curling and opening claspers, 4) copulation. Bioassay results demonstrated that female body-wax lipids elicit a sex pheromonal activity in male flies. The active substances were located on all parts of the female body. Teneral females showed a potential to evoke mating responses from males and males responded even 30 min after eclosion. Effects of light intensity on *B. paupera* male mating behavior and upwind flight to pheromone source were assessed in cage and wind tunnel tests. Typical mating behaviors were not affected by light intensity. However, male upwind flight to the pheromone source was remarkably inhibited by low light intensity, which significantly depressed their flight activity. n-aldehydes (C7-C18), n-hydrocarbons (C13-C22) and 2-ketones (C16-C18) were the main components of female body-wax lipids. Several GC-EAD active components in the extracts were separated by preparative capillary-GC, followed by GC-MS analysis. Four n-aldehydes (C10-C13) showed prominent activity to males in cage assays. 2-ketones and n-hydrocarbons showed none or lower activity. Sex pheromonal activity of these aldehydes was also confirmed by trap tests in a mushroom cellar.

Evidence for chemical mediation of host range expansion & host shifts in papilionid butterflies

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Phytochemical constraints are thought to be one of the major determinants of host range evolution and host shift in phytophagous insects. Papilionid butterflies comprising over 560 species utilize, for larval growth, a limited range of plants in the families Aristolochiaceae, Annonaceae, Lauraceae, Magnoliaceae, Rutaceae or Apiaceae. Although a number of oviposition stimulants have so far been reported for papilionids, very little is known about phytochemicals that reasonably explain how they have accomplished drastic host shifts. We identified oviposition stimulants for a Rutaceae feeder, *Papilio polytes,* from *Toddalia asiatica, Citrus depressa*, and *Glycosmis citrifolia*, and also for a Magnoliaceae feeder, *Graphium doson*, from *Michelia compressa* and *Magnolia grandiflora*. Two compounds (*trans*-4-hydroxy-*N*-methyl-L-proline and 2-*C*-methyl-D-erythronic acid) identified from *T. asiatica* and *G. citrifolia,* and five compounds (stachyine, proline, quinic acid, synephrine, and choline) identified from *C. depressa* were regarded as important constituents which synergistically stimulate egg-laying by *P. polytes.* On the other hand, pinitol, identified from *M. compressa* and *M. grandiflora,* was found to be one of the key stimulants responsible for host recognition by *G. doson*. Based on our findings and previous data, we discuss a possible phytochemical-mediated pathway of host range expansion and host shifts in papilionids.

The repellency of hinokitiol against the cigarette beetle, *Lasioderma serricorne*

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The repellency of 73 chemicals against the cigarette beetles was investigated with an olfactometer. Hinokitiol strongly repelled the beetles. The repellency of hinokitiol was evaluated in a semi-field test. The filter-paper tapes treated with hinokitiol were attached around the openings of the cardboard boxes containing cured tobacco. The hinokitiol tapes prevented the beetles from entering the boxes. However, in the olfactometer assay, the filter paper treated with hinokitiol did not show repellency after 48 hr. Hinokitiol sublimed rapidly and its 90% sublimed within 48 hours. Therefore, a repellent tape slowly releasing hinokitiol was developed. The tape consisted of a filter paper containing hinokitiol, a polyethylene film bag sealing the paper, an aluminium sheet on the backside of the bag and an adhesive tape on the aluminium sheet. The tape slowly released hinokitiol. In the semi-field test, the tapes attached around the openings of the cardboard boxes containing food attractant prevented the beetles from entering the boxes. The number of the beetles in the treatment boxes was less than 20% of that in the control boxes for 64 days. The effectiveness of the tape continued for 92 days.

The effects of dynamic experience of resource heterogeneity on oviposition behavior of the seed beetle, *Callosobruchus maculatus* (F.)

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The effects of dynamic search experience in response to resource heterogeneity and its depletion on oviposition behavior of the seed beetle, *Callosobruchus maculatus*, are studied here through a longitudinal view and a new set of experimental design. Sixty-four adzuki beans, 16 from each of four sizes, were arranged into three designs of 2 ´ 2, 4 ´ 4, and 8 ´ 8 sub-lattices of increasing resource heterogeneity, thus making the dynamic experience of size change by the female significantly different. We assumed each bean size has equal encountering probability. The results showed that, the difference between proportions of eggs laid on large beans and small ones increased with increasing resource heterogeneity. While the female still maintains the egg-discrimination as the major host selection trait, we show that the trait of preferring larger hosts is maintained for the entire oviposition process only if enough stimuli of contrasting experience of resource heterogeneity are retained in the environment. These results imply that the seed beetle does compare bean quality experience in the information processing constituting her decision-making capability. We also tested whether the size information content employed by the female is the weight of the host bean instead of the surface-area.

Trail-pheromone perception in the eastern subterranean termite

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The chemo-receptors responsible for trail-following behavior in *Reticulitermes flavipes* was investigated using behavior choice tests. The trail following behavior of worker, soldier, prealate, and young casts was examined as well. Termites were anesthetized on ice before being manipulated one of the six potential receptors, including antennae, labial palps, maxilla palps, and tarsi of pro-, meso-, and meta-thoracic legs. Results show that only termites with antennae manipulated showed no response to trail-following mimic, indicating the chemo-receptors are located mainly, if not solely, on antennae. Further tests revealed that only the distal ten flagellar segments perceive the trail-following substances. Additionally, there was a strong linear relationship between the number of flagellar segments removed and the time required to locate the trail and the distance traveled on the trail. All the four castes demonstrated trail-following behaviors. However, worker and prealate tended to locate the trail faster than soldiers and young. Interestingly, both worker and young displayed reversed trail-following, suggesting either recruitment behavior or reinforcement of the trail.

Seasonal changes in metabolic rate & water loss through the spiracles and cuticle in the goldenrod gall fly, *Eurosta solidaginis* **(Diptera, Tephritidae)**

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The goldenrod gall fly, *Eurosta solidaginis*, overwinters as a diapausing third-instar larva within its gall on the stem of goldenrod (*Solidago* spp.). During the winter this is a severely environment and the diapausing larvae are highly resistant to water loss. We used flow-through respirometry to track decreases in metabolic rate and water loss (cuticular and spiracular) as the larvae moved into winter diapause. The anterior spiracles were more important to respiration as there was no change in metabolic rate or water loss when the posterior spiracles were blocked. Conversely, blocking the anterior spiracles caused CO_2 production to decrease by almost 50%, although the rate of water loss fell only slightly. The rate of water loss fell as larvae moved into diapause because of a decrease in metabolic rate and a reduction in the permeability of the cuticle. We conclude that there is a substantial drop in the rate of water loss and metabolic rate as the goldenrod gall fly enters diapause in the autumn. A high cuticular resistance to water loss and efficient spiracles contribute to this species' notably low rate of water loss.

Host plant orientation and feeding in the rice bug, *Leptocorisa chinensis* **(Dallas) in response to olfactory cues**

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The rice bug, *Leptocorisa chinensis* Dallas (Hemiptera: Alydidae) causing pecky rice is one of the most important pests in Japan. It has been difficult to investigate chemicals affecting its behavior because no proper bioassay systems were available. We developed a bioassay method using a two-choice test to assess the response of *L. chinensis*, nymphs to olfactory stimuli. Two plastic cups were placed on a board covered with wet quartz sand. Each of the cups was covered with mesh cloth in order to prevent visual stimuli of the odor sources tested. At the center of the board, a stick was erected to function as the releasing point for bugs tested. This two-choice test bioassay method was used to demonstrate the responses of *L. chinensis* nymphs to olfactory stimuli. As a result, host plant volatiles alone were not attractive to the bugs, whereas the number of responding bugs significantly increased when there were feeding nymphs of *L. chinensis* together with host plants. We also developed a bioassay method to assess feeding response of *L. chinensis* to host plant extracts.

Herbivore-induced volatiles from *Acer negundo* **and their defensive function against** *Anoplophora glabripennis*

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Ashleaf maple (*Acer negundo*) is the highly preferred host of adult *Anoplophora glabripennis* Motsch (ALB). However, what will happen after ALB's feeding-damage? Attract or repel? We investigated the behavior responses of ALB to the feeding-damaged *Acer negundo* over post-infestation duration by Y-tube olfactometer. The data indicated that the intact *A. negundo* attract the ALB evidently, but the infested plant repelled the ALB significantly from 24h to 48h after feeding damage. The volatiles from the infested plant were analyzed by TCT-GC/MS. It showed that many chemicals were induced, and the emission amount of most volatiles reached the maximum at 48h after infestation, then reduced sharply at 72h. The behavior response data of ALB to individual artificial compounds showed the herbivore-induced volatiles, such as Ocimene, trans-Geranylacetone, 2-Methylbutyronitrile, syn-2-Methylbutylaldoxime, 1,3,3-Trimethyl-2-methylene-indoline, repelled the ALB intensively. The peak time of releasing amount of herbivore-induced volatiles were well matched with the time of high repellency to ALB, which well exhibited the defensive function of infested ashleaf maple to prevent the further attack from ALB, at least within 72h. The mechanical damaged ashleaf maple was investigated simultaneously which showed the similar repellent trend but less number of volatiles be induced.

Disruption of pheromone communication in *Choristoneura rosaceana* and *Pandemis limitata* (Lepidoptera: Tortricidae) using microencapsulated single and multi-component pheromone blends

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Microencapsulated (MEC) pheromones are being developed to disrupt mating of Tortricid pests of pome fruits in western North America. Responses of male *Choristoneura rosaceana* (Harris) and *Pandemis limitata* (Robinson) to calling females in clean air, and air treated with their major pheromone component, (*Z*)11-tetradecenyl acetate (*Z*11-14:OAc), released from 3M Phase I microcapsules applied at doses of 1, 10 and 100 mg ai m^{-2} to a flight tunnel (= field rates of 10, 100, and 1000 g ai ha^{-1}) were compared. Disorientation in both species was dose-dependent. Relative to orientation to calling females in clean air, orientation of male *P. limitata* was disrupted 23.3, 46.3 and 71.3%, and orientation of *C. rosaceana* was disrupted 31.6, 37.7, and 45.8% by doses of 1, 10, 100 mg m^{-2}, respectively. Disruption lasted 26, 74, and 218 h in *P. limitata* and 30, 54, and 174 h in *C. rosaceana* at these doses, respectively. Disruption of *P. limitata* was greater than *C. rosaceana* and appeared correlated with species' differences in female pheromone release rates. Multi-component blends caused greater disruption in *P. limitata* than the main component, but only at the lowest application rate. Disruption mechanisms invoked by MEC pheromones are discussed.

Female sex pheromone & communication disruption in the rice leaf bug, *Trigonotylus caelestialium* (Kirkaldy) (Heteroptera: Miridae)

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The rice leaf bug, *Trigonotylus caelestialium*, injures rice, wheat, maize, and gramineous forage grasses. Females attract conspecific males (Kakizaki and Sugie, 1997), and the female sex pheromone was identified as a mixture of *n*-hexyl *n*-hexanoate (H:H), (*E*)-2-hexenyl *n*-hexanoate (*E*2H:H), and *n*-octyl *n*-butyrate (O:B) by the GC-EAD method and GC-MS analyses. H:H and *E*2H:H are essential components in the sex pheromone, and O:B enhances the male attraction in combination with the essential 2-components (Kakizaki and Sugie, 2001). The suppression effects of *T. caelestialium* populations by communication disruption using the sex pheromone were examined. In small-scale field tests (100 m^2), the numbers of males captured by traps baited with a lure containing the sex pheromone were reduced to some degree by treatment with either H:H, *E*2H:H, or a 2-component mixture, furthermore, those baited with the lure or 3-virgin females were mostly reduced by the 3-component treatment. In large-scale field tests (10,000 m^2), the population densities and males catches by traps in the treated fields were lower than those in the untreated fields. However, the traps baited with females had slightly attraction activity in the treated field, further, it may indicate that unknown pheromone components are involved with male attraction processes.

Effective pheromone lures for monitoring the rice leaf folder moth, *Cnaphalocrocis medinalis* (Lepidoptera: Crambidae)

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Several factors affecting the attractiveness of traps baited with synthetic sex pheromone components of *Cnaphalocrocis medinalis* were examined in the laboratory and field. In the laboratory, among the four pheromone components, Z11-18: Ald, Z13-18:Ald, Z11-18:OH and Z13-18:OH, the major component, Z13-18:Ald, was shown to be an essential component for attraction and Z11-18:Ald was shown to be a potent synergist for the major component. The alcohols also showed an apparent synergistic effect in the laboratory bioassay when their mixture was combined with an aldehyde mixture. The addition of the geometric isomers of the four components at levels greater than 5% of the purified 4-component blend showed significant inhibitory effects on the pheromonal activity in the laboratory. In the field, no synergistic effect of the alcohols on the aldehydes was shown. A range of loadings of the pheromone rubber septa (0.01–3.42 mg) was tested and all remained attractive for at least one month with optimum attractiveness on 0.86 mg/septum. No significant effects were obtained when the ratio of Z11-18:Ald to Z13-18:Ald was changed. Rubber septa containing 0.86 mg of the four components (*E*-isomers 5%) at the natural ratio are being effectively used for monitoring.

An artificial blend of volatiles for attracting parasitic wasps

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When damaged by herbivorous arthropods, plants are known to emit specific volatile compounds that attract the natural enemies of herbivores. If we can artificially mimic particular blends of volatiles, we may be able to use them in agroecosystems to enhance the effectiveness of pests' natural enemies. The objective of this study is to test this possibility in a tritrophic system consisting of cabbage plants, diamondback moth larvae (*Plutella xylostella*) and their specialist parasitoids, *Cotesia plutellae*. The specific volatile components emitted from cabbage plants infested by diamondback moth larvae were analysed using a gas chromatograph-mass spectrometer (GC/MS). By using a choice-chamber bioassay, we found that a mixture of four compounds (hereafter called blend) attracted the wasps. The blend was significantly attractive to parasitoids over a certain range of doses, but it was not attractive at higher and lower doses. We dissolved the blend in triethyl citrate, and added the solution to a glass instrument from which the blend was gradually vaporized in the air. By using this equipment, we tried to attract the parasitic wasps under both greenhouse and field conditions. Possible applications of the blend in agroecosystems will be discussed.

Sex pheromone composition of the diamondback moth, *Plutella xylostella* in Korea

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The diamondback moth (DBM), *Plutella xylostella,* is one of the most widely distributed moths, and the most destructive pest on cruciferous crops throughout the world. Three components of DBM sex pheromone has been known: (*Z*)-11-hexadecenal (*Z*11-16:Al), (*Z*)-11-hexadecenyl acetate (*Z*11-16:Ac) and (*Z*)-11-hexadecenyl alcohol (*Z*11-16:OH). The pheromone gland extracts of female DBM in Korea were analyzed with Gas Chromatography (GC). Three components from DBM sex pheromone were identified: the highest peak was *Z*11-16:Al and the other two components also detected. To examine DBM male response to female sex pheromone, Electroantennogram (EAG) assay to the synthetic sex pheromone components of DBM and their analogues was done. In EAG assay the two sex pheromone compounds, *Z*11-16:Al and *Z*11-16:Ac, elicited significant antennal response, but the response to the other compounds tested was weak, except for *Z*11-14:Ac and *Z*11-16:OH. In blend assays, the blend of Z11-16:Al and Z11-16:Ac showed strong response at the ratios of 5 : 5 to 7 : 3. In field trapping tests conducted at Chinese cabbage fields in Korea, DBM males were attracted mainly to 7: 3 blend.

The effects of photoperiods on the daily locomotion pattern of the German cockroach, *Blattella germanica* (L.)

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The locomotion of the German cockroach, *Blattella germanica* (L.) can be entrained by light dark (LD) cycles, but the following constant darkness (DD) can cause a free-running rhythm on the locomotion. When cockroaches were entrained to various light-dark regimes, they showed typical locomotor pattern of nocturnal insects by concentrating their locomotion in the scotophase of the photoperiod. When the scotophase became extremely short (2 hours long), the locomotion pattern did squeeze in but the pattern was quite different from the one that was exposed to longer scotophase. Interestingly, the locomotor patterns did change during free-running condition after exposing to the different Zeitgebers. This change only showed in the distribution pattern of locomotion rather than in the length of circadian period, because all the Zeitgebers were 24 hours. By superimposing the locomotor distribution patterns of the same individuals under DD following two different Zeitgebers along the circadian time, the after effect was shown in the phase shifting. The locomotion pattern was also analyzed to understand the interaction between the circadian clock and locomotion.

Effects of herbivore-induced plant volatiles & previous foraging experiences on the residence time of the predatory mite *Amblyseius womersleyi* in a prey patch

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It is important for predators to make optimal decisions on how long to stay in a prey patch. We investigated the effects of herbivore-induced plant volatiles, previous foraging experiences of predators, and the current patch condition on the residence time of *Amblyseius womersleyi*. The longest residence time was observed when the predators were in a heavily infested-patch. Neither prey eggs alone nor herbivore-induced plant volatiles alone emitted from the currently inhabiting patch affected the residence time. Satiated predators that had previously experienced starvation stayed longer in a patch than those that had not experienced starvation. When *A. womersleyi* had previously experienced a prey-rich patch, it showed approximately the same residence time irrespective of the number of prey in a patch. By contrast, *A. womersleyi* that had previously experienced a prey-poor patch stayed longer in a patch of high prey density than in one of low prey density. The residence time was also affected by herbivore-induced plant volatiles from outside the prey patch. *A. womersleyi* stayed significantly shorter in the volatiles-airflow than in the control-airflow. Thus, *A. womersleyi* appears to determine its residence time based on these multiple information about its foraging habitats.

Foraging & energy balance in *Pachydiplax longipennis* (Odonata: Libellulidae): implications for reproductive success

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Food intake, flight duration, and an estimate of prey availability at feeding areas were quantified for the dragonfly, *Pachydiplax longipennis*, by observing focal individuals on artificial perches. From these data we constructed activity budgets and thence have calculated approximate energy budgets. The latter indicate that average energy costs of foraging are small, and most individuals are in positive short-term energy balance. Nevertheless, these dragonflies probably have to feed 9-14 h per day to maximize reproductive success, and energy requirements may limit the time that males can spend at mating territories. Because of the energy requirements of territory defense by males and egg production by females, energy demands on both sexes appear to be high and roughly similar. Consequently, males may be relatively risk prone in foraging owing to high variance in male reproductive success.

Ecophysiological screening of necrotizing spiders in Korea using nonradioactive sphingomyelinase assay

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Spider bites cause a range of symptoms from simple swellings to disfiguring necrotic lesions, and occasionally death. There are now more than 40,000 identified spider species in the world, and considered about 100 species as actually dangerous to human. While spider bites are not a major medical problem in Korea, it would be of great value to know which species of spiders pose a threat to human health. A middle molecular weight protein, sphingomyelinase D, has been identified in the venom of the brown recluse spider and strong evidence suggests that they have a major role in spider bite necrosis (Tambourgi *et al.*, 1998). For the identification of necrotizing species, we have investigated using recently developed non-radioactive assay of sphingomyelinase for rapidly screening the necrotizing venoms. Here, we demonstrate the fetal toxicity of 47 species (34 genera within 15 families) among 622 identified spider species of Korea. It has been revealed that one species of the orb-weaving spider, *Araneus ventricosus*, and another species of wandering spider, *Dolomedes sulfureus* have the strongest positive activities among themselves.

Long chained acetates in the defensive secretion of Thysanoptera

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Defensive secretions of several Thysanoptera species were found to contain mixtures of long chained acetates: saturated decyl, dodecyl, tetradecyl, hexadecyl and octadecyl acetate. If the saturated acetates are solid they are mixed with liquid long chained saturated alkanes, e. g. pentadecane. A further method is to dissolve solid saturated acetates in unsaturated acetates, e. g. (11Z)-11,19-eicosadienyl acetate. Those mixtures spread fast on surfaces and are especially designed to produce a long-lasting coating of sensory organs of potential predators like ants and mites. Furthermore unsaturated acetates act as repellents against ants. Similar mixtures, e g. (11Z)-11-eicosenyl acetate with octadecyl acetate, can be found in Thysanoptera and in some other insects species as well.

Secondary metabolites from sunflowers as oviposition stimulants for the banded sunflower moth, *Cochylis hospes*.

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The banded sunflower moth, *Cochylis hospes* (Walshingham) (Lepidoptera: Cochylidae) (BSFM), is a specialist species on sunflowers (*Helianthus* spp.). Its larvae are restricted to feeding on sunflower heads and are a significant pest of cultivated sunflowers in the USA. Extracts of sunflower head surface chemicals stimulated oviposition by *C. hospes* when applied to an artificial substrate in an egg-laying bioassay. The major compounds in these extracts were epicuticular waxes (long chain aldehydes and alcohols) and monoterpenes, primarily α-pinene. Also present were a number of sesquiterpenes, sesquiterpene lactones, and diterpenoid acids and alcohols previously characterized in studies of *Helianthus* secondary metabolites. Fractionation of head extracts by column chromatography and HPLC resulted in the isolation of several compounds which stimulated oviposition by females in the bioassay, and are apparently responsible, at least in part, for host acceptance by female *C. hospes*. Chemical characterization and indication of the final purity of active compounds used in bioassays was carried out by GC-MS and NMR spectroscopy.

Feeding responses of a dampwood termite, *Zootermopsis nevadensis* to various wood extracts

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A dampwood termite, *Zootermopsis nevadensis* **(Hagen) is regarded to be introduced into Japan with imported timbers from North America. It locally distributes in Kansai region and inhabits in fallen pinewoods (***Pinus densiflora* **(Sieb et Zucc.)). In the present study, we focused the feeding preference of** *Z. nevadensis* **to the sapwood extracts and investigated the behavioral and electrophysiological responses to the extracts of five softwoods and one hardwood (***P. densiflora*, *Picea jezoensis* **Carr.,** *Abies sachalinensis* **Fr.Schm.**, *Larix leptolepis* **Gord.,** *Cryptomeria japonica* **D.Don,** *Fagas crenata* **Blume). According to the two-choice feeding tests,** *Z. navedensis* **tended to prefer in this order,** *A. sachalinensis> C. japonica*, *P. densiflora, P. jezoensis, F. crenata > L. leptolepis*. **Contact chemoreceptor neurons in taste hairs on the labial and maxillary palps generate less adaptive impulses to these extracts. However, parallel relationships between the behavioral and electrophysiological results would not be clear. Probably, different cells were fired by different extracts, and/or olfactory stimuli may effect on the feeding preference of** *Z. navedensis*.

Electrophysiological & behavioural activity of chocolate volatiles: attractants for the pyralid moths *Ephestia cautella* **&** *Plodia interpunctella*

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The use of food odours to monitor females in pest populations would be a valuable complement to pheromone-based methods. In a chocolate factory stored product moths were observed flying in the vicinity of containers with chocolate-based products. We tested three chocolate-based products in flight tunnel experiments and found that they all attracted males and mated females of the pyralids *Ephestia cautella* and *Plodia interpunctella.* The substances in chocolate-based products were isolated by two different extraction methods to; steam distillation and head space collection. GC-EAD experiments revealed several electrophysiologically active substances in the different products. The extracts and headspace samples were analysed with GC-MS and the substances identified. The electrophysiological activity of the corresponding synthetic samples was investigated by EAG, and two components with a positive dose-relationship were detected for both species and sexes, phenylacetaldehyde and nonanal. These two compounds were also behaviourally active when tested individually in a flight tunnel, as was a third substance, ethyl vanillin. A blend of the three components induced landing rates corresponding to the levels found when testing chocolate-based products, suggesting that we had found the chemicals mediating the attraction to chocolate. Field tests with the blend are now running to evaluate the potential use in monitoring traps.

Oviposition behavior of *Stenoma catenifer* **Walsingham: chemical & physical stimuli & time of egg-laying**

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The influence of chemical and physical stimuli on the oviposition behavior of *Stenoma catenifer* Walsinghan (Lepidoptera: Elachistidae), as well as time of egg-laying were investigated aiming for biological control studies with *Trichogramma* spp. The experiments were carried out in the laboratory (25±2ºC temp., 60±20% RH and 14-h photophase), using 5 couples per cage. To evaluate the chemical stimulation, the cages were internally coated with paper towel with the different treatments: 1 and 2) fruits of cultivar Margarida and Breda; respectively 3) artificial fruit; 4) fruitless (natural or artificial). In order to evaluate the physical cues we used hollowed paper towel, non-hollowed paper towel and a smooth sheet of paper. The number of eggs in all treatments including the avocado fruit was statistically higher than the remaining ones. Likewise, the number of eggs on the hollowed paper towel was statistically higher in comparison with the non-hollowed paper towel and smooth sheet of paper. The peak of egg laying in laboratory was from 8PM to 12PM. Thus, we verified that achieving eggs on *S. catenifer* required coating the cage with a hollowed paper towel (physical cue) including the avocado fruit (chemical stimulation).

Biochemistry & evolution of floral scent

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Floral scents of plants are often complex mixtures of volatiles derived from several metabolic pathways. The number of presently known volatile compounds from flowers is approaching a thousand, and new compounds are constantly being discovered in various plant species. Closely related species have distinct volatile profiles that can be discriminated by specialized insect pollinators, thus limiting gene flow across species boundaries. The ability of related plant species to synthesize different volatiles in their flowers is sometimes due to differential gene expression. However, new enzymes that catalyze the formation of new floral scent components are constantly evolving through the process of gene duplication and divergence, allowing for the majority of floral scent divergence. It has been shown that very small changes in protein sequences can lead to the ability of the changed enzymes to catalyze the transformation of drastically different substrates, leading to the production of new floral scent compounds. Such evolution is facilitated by the presence of large gene families in plant genomes that already encode similar enzymes with slightly different substrate specificities; the large number of such genes makes genes duplications even more likely to occur.

The glucosinolate-myrosinase defence system in the cabbage aphid, *Brevicoryne brassicae*

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Cruciferous plants contain glucosinolates that are hydrolysed by myrosinase enzymes within the plant to produce volatile isothiocyanates which act as repellents for generalist pests but attractants for specialist crucifer feeders including the cabbage aphid, *Brevicoryne brassicae*. Specialist aphids also produce a myrosinase enzyme and assimilate high levels of plant glucosinolates. However, the biological role of the aphid enzyme has not been explored. It is likely that the aphid myrosinase and sequestered glucosinolates play a defensive role against predators and parasitoids. Aphid myrosinase is found only within muscle tissue of the head and thorax while the glucosinolates are most likely located in the haemolymph, a spatial arrangement suggestive of a defensive role. Recent research has concentrated on the dynamics of the glucosinolate-myrosinase system in aphids, particularly glucosinolate uptake, released volatiles and the biological role of this defensive system in *B. brassicae*.

Evidence of female sex pheromone in *Anoplophora versteegi* Ritsema (Coleoptera: Cerambycidae) – a serious pest of citrus in north eastern hill region of India

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Severe epidemics of trunk borer *Anoplophora versteegi* Ritsema (Coleoptera: Cerambycidae) were noticed on mandarin (*Citrus reticulata* Blanco) in the North Eastern Hill Region of India for the last 15 years, which resulted in gradual decline of mandarin orchards to the extent of 10-15% every year. The olfactometer studies revealed that the extract from lower abdomen of female beetle possessed pheromone properties and attracted 100% adult males within 5 minutes when kept at 5 meters apart. The pheromone continued to attract males for more than 7 days in laboratory conditions. The extracted material kept at 20 meters apart in a control orchard increased the activity of male beetles on surrounding citrus trees. When antennae of the male beetles were removed, they continued to be attracted towards females and copulation took place indicated that males used visual cues in the absence of antennae for locating adult females. The pheromone extracted from gravid females was also found to attract the male beetles to a lesser extent evidenced by mating between males and gravid females. The chemistry, structure, activity and relationships of the pheromone were under progress. This is the first report about the presence of female sex pheromone in *A. versteegi* from India and also seems to be first in the world.

Expression & properties of ant apolipophorin-III

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Apolipophorin-III (apoLp-III) is present at relatively high levels in the antenna of the male red imported fire ant, *Solenopsis invicta* (Formicidae: Hymenoptera). PCR analysis shows that apoLp-III is expressed in the male antenna as well as in other male body segments, and it is also expressed in workers. ApoLp-III is known to be involved in transport of diacylglycerol through the hemolymph from fat body to flight muscle in locusts. However, male fire ants do not have a high fat content and probably do not use fat as a major fuel for flight. Additional known functions for apoLp-III include immune response and apoptosis, and lipophorins also carry cuticular hydrocarbons and pheromones in some insects. Therefore, we are investigating whether apoLp-III might have an olfactory function in fire ants. We expressed fire ant apoLp -III in *E. coli* and the purified recombinant protein was tested for hydrophobic ligand-binding properties, using the fluorescent dye ANS. We found that ANS binds to fire ant apoLp-III with a dissociation constant in the micromolar range, similar to ANS binding to intracellular lipid-binding proteins. We are currently testing the capacity of fire ant apoLp-III to bind endogenous ligands.

Host finding cues of *Platygaster demades*, parasitoid of apple leaf curling midge & pear leaf curling midge

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Platygaster demades is an egg parasitoid of the apple leaf curling midge (ALCM), *Dasineura mali* and pear leaf curling midge (PLCM), *Dasineura pyri*. The response of one to two day old naïve female parasitoids to pear and apple plant odour cues was measured in a specially designed two armed Y-tube olfactometer. When the parasitoids were given a choice of odours of ALCM and PLCM egg-free apple or pear leaves or clean air, they preferred the plant cues, responding similarly to both apple and pear odours. The results indicate that *P. demades* utilises plant cues to locate the habitat of its host and then searches for host eggs to parasitise. The odour mediated responses to midge egg-free apple and pear leaves suggest that the odours are not host induced and also that *P. demades* is attracted to a cue common to both apple and pear leaves.

Diffraction gratings in insect cuticle & spectral iridescence in Coleoptera

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Iridescence in beetles is usually due to interference, in which light is scattered as it passes through cuticular layers of varying density. In a few families, however, iridescence is due to diffraction gratings formed by cuticular microsculpture. In these unusual taxa, patches of spectral iridescence are visible under incident light, usually at the elytral apices. Cuticular diffracting structures vary widely in form; some resemble flattened setae, while others take the shape of minute combs or tabs perpendicular to the surface of the cuticle. This iridescence may be a form of crypsis, an intraspecific cue, or may simply be an artifact of the plastron-like pubescence related to moist environments. This phenomenon has hitherto been observed only in Scarabaeidae, Carabidae, Gyrinidae, Phalacridae and Torridincollidae; recently, several iridescent species in Hydrophilidae and Leiodidae were discovered. In order to elucidate the evolutionary origins and possible functions of these diffraction gratings, representatives of each family were examined with both light photography and scanning electron microscopy. Cuticular diffraction gratings appear to be linked to moist or aquatic habitats, but are likely visual cues as well; most of the iridescent species are diurnal and occur near the equator, where the angle of incident light is very low.

Evolution of pheromone desaturases in primitive moths (Lepidoptera)

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Typical moth sex pheromones like unsaturated 12-, 14- or 16-carbon acetates and alcohols are produced in the female pheromone gland from palmitic/stearic acid that is modified in a few enzymatic steps: desaturation, oxidation and reduction. The diversity of pheromone components in different species is obtained by the specificity of the enzymes involved and from the reactions occuring in different order. In the primitive moths and in the sister group Trichoptera short-chain alcohols and ketones are used as pheromones, instead of the long-chain pheromones used in the rest of Lepidoptera. *Lampronia capitella* (Incurvariidae) is the most primitive moth for which a palmitic/stearic acid-derived pheromone is known whereas the primitive moth *Eriocrania semipurpurella* (Eriocraniidae) use a short-chain pheromone similar to Trichopteran pheromones. Based on DNA-sequences of pheromone desaturase-genes in *Eriocrania semipurpurella* and *Lampronia capitella* together with previously described desaturase-genes from higher moths, we have made a phylogenetic reconstruction of the evolution of the genes and describe how the pheromone production has evolved from the primitive to the more recent Lepidopteran species.

Multiple mating stimulates the oviposition but not the egg maturation in female seed beetle (*Callosobruchus maculatus* (Fabicius))

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In general females, compared to males, do not enhance reproductive success by mating multiply; however, an evolutionary inclination to mate multiply in females has been observed in a variety of insect species. In this study, we tried to determine the potential function of additional matings for female *Callosobruchus maculatus* by permitting them to mate either singly or doubly. The lifetime fecundity and weight loss were both higher in the doubly than singly mated females, while the longevity was not significantly different between treatments. In addition, there was an immediate increase of egg production on the remating day of doubly mated females. The dissection of ovaries carried out 24 hours after remating showed that the number of eggs unlaid was lower in doubly mated females, thus multiple mating did not directly stimulate egg maturation. Indirect driven hypothesis that states the increased egg maturation rate was driven under the approximate emptiness of ovaries was proposed. By depriving hosts of females for 3 days after remating to inhibit oviposition, we were able to support this hypothesis because the increase of egg production disappeared in doubly mated females due to the number of eggs unlaid remaining high. Therefore, in *C. maculatus*, multiple mating for females could contribute to reproductive success through direct effect of increasing ovisopistion and indirect effect of increasing egg maturation.

Effects of prey species & prey densities on the olfactory responses of a generalist predator to herbivore-induced plant volatiles (HIPV)

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To clarify the effects of prey species and prey densities on the long-distance prey finding by a generalist predator *Wollastniella rotunda*, we studied the olfactory response of the predators toward HIPV emitted from eggplants infested by thrips *Thrips palmi* versus those from eggplants infested by spider mites *Tetranychus kanzawaii* in a Y-tube olfactometer. When the number of each prey species was low per plant (200 individuals each), the females preferred the plants infested by *T. kanzawaii* over those infested by *T. palmi*. By contrast, when the number of each prey species was high (800 individuals each), the females showed the equal distribution between the two HIPV sources. When the number of prey per plant was different in two prey species, the females significantly preferred HIPV from plants infested by prey of high-density regardless of prey species. These results suggest that the effects prey densities on the quality and/or quantity of HIPV differ according to herbivore species, and the predator females would distinguish such differences to select the plants with higher prey densities. This behavior would increase the foraging efficiency of the generalist predator.

Role of leaf volatiles from two maize (*Zea mays* L.) cultivars in eliciting larval orientation of *Chilo partellus* (Swinhoe) (Lepidoptera: Pyralidae)

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The effects of two maize cultivars, susceptible (Basilocal) and resistant (Kisan), and their methanol- and hexane- extracts in eliciting orientation of the first-instar *Chilo partellus* (Swinhoe) larvae were studied in the laboratory. When presented at one end of the orientation tunnel, leaf whorls of both the Basilocal and Kisan varieties, elicited higher orientational responses of stemborer larvae as compared to the blank end, the orientation preference (OP) of the larvae being + 72% and + 48%, respectively. Statistically, OP of larvae for Basilocal leaves was significantly higher than for Kisan leaves ($P<0.01$). Methanol extracts of both the Basilocal and Kisan leaves also attracted the neonate larvae, but the difference between the OP for the two was not significant. However, the attraction of larvae for hexane extractable chemicals of Basilocal (OP= 40%) was significantly higher than that for Kisan (OP = -32%). The observations indicate that difference in the orientational response of stemborer larvae for the two maize cultivars is due to non-polar semiochemicals, extractable in hexane. Identification of the chemicals in these maize cultivars can be useful in the behavioural management of this economically important insect pest of maize and sorghum.

Host plant preferences of diamondback moth (*Plutella xylostella* L.) in western Australia.

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In Western Australia, diamondback moth (*Plutella xylostella* L.) has become a serious problem in canola (*Brassica napus*), having repeatedly reached outbreak levels in some regions. At present, the factors responsible for *P. xylostella* outbreaks in canola are poorly understood. Field surveys in Western Australia have established that *P. xylostella* is abundant on some weedy Brassicaceae, indicating that alternative hosts may be contributing to the problem on canola. Two experiments were conducted to assess the host plant preferences of gravid female diamondback moth when offered a number of Brassica host plants. The purpose of these experiments was to determine whether *P. xylostella* actively prefers canola over weedy or horticultural hosts, and whether this preference results in a larger egg loads on canola. The first experiment used a Y-tube airflow olfactometer to assess the immediate behavioural response of *P. xylostella* to volatiles from a range of host plants. The second experiment assessed the oviposition preferences of the moths when exposed to the same set of host plants over an extended time period. The results of the experiments were then compared to assess whether the oviposition preferences of *P. xylostella* reflected the observed olfactory responses.

When did the discovery of olfaction communication take place?

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It is generally agreed that the first published observations about communication between insects by olfaction were the fact of the French entomologist Jean-Henri Fabre, at the beginning of the 20th century. In fact, we discovered mentions of olfaction communication in the works of Charles Bonnet and Louis Jurine, two well-known 18th century entomologists from Geneva Switzerland. Bonnet noted that ants always follow the same routes. Rubbing that route with a finger interrupts it. Bonnet hypothesized that ants deposit a secretion which is perceived by olfaction. He so discovered the trail pheromones. Jurine reported, and noticed himself, that some great female moths attract males at long distances. The place where the female was is attractive even in the absence of the female. Jurine again was hypothesising an odour evaporated by the female, which attract the males by olfaction. Depriving males of their antenna do not allow them to find the females. The publications of these observations took place in 1779 for Bonnet, in his book on "Observations sur les Insectes". Jurine placed his account about moths in the beginning of his great book "Nouvelle manière de classer les Hyménoptères", published in 1807.

Assessing the pitfalls of insect repellent evaluation methodologies

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Insect repellents provide useful protection from malaria, for high-risk groups including forest-workers; especially in the Amazon Basin where vectors are difficult to control through conventional means due to their exophily, crepuscular biting and forest-breeding. The WHO recommends that repellents are field-tested using human-landing catches, with individual collectors positioned >10m apart. However, it is still commonplace to perform tests using repellent treated "bait" and an unprotected "collector". This could overestimate repellent protection and longevity since mosquitoes will feed preferentially from the unprotected "collector", thus potentially exposing subsequent users to disease. Mosquito diversion from 15% deet (repellent) to 15% baby oil (control) was quantified utilising pairs of testers with each team situated >10m apart. Permutations of repellent repellent; control control; and repellent control were replicated over 18 nights with 6 pairs. Control individuals received 22.82% more bites when paired with repellent wearers, than untreated individuals (likelihood-ratio test Prob > chi2= 0.0485), and repellent wearers received 11% more bites when paired with other repellent wearers than controls (N.S.). In a further test with 25% lemongrass *(Cymbopogon citratus)* repellent, controls received 23.53% more bites when paired with repellent wearers, than with unprotected partners (N.S.). Thus it is recommended that repellents are field-tested using individuals and not pairs of collectors.

Assessing the pitfalls of insect repellent evaluation methodologies

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Male-produced anti-sex pheromone in a plant bug.

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In at least some plant bugs (Miridae) females produce sex pheromones in the metathoracic scent gland (MSG), which in most other true bugs (Heteroptera) is responsible for chemical defense. Females of a pine inhabiting mirid, *Phytocoris difficilis* Knight, contained predominantly hexyl, (*E*)-2-hexenyl and (*E*)-2-octenyl acetates in their MSG, while the latter two compounds were found only in trace amounts in males. This ternary acetate blend (2: 1.5 :1 by volume) constitutes the sex pheromone of *P. difficilis.* However, the possibility that the MSG secretion of male plant bugs plays a role other than defense has been largely overlooked. For *P. difficilis* we showed that hexyl butyrate and (*E*)-2-hexenyl butyrate are abundantly produced only in males, and demonstrated that these MSG compounds elicit strong antennal responses in conspecific males. Furthermore, physiologically relevant doses of these butyrates totally interrupted attraction of males to the female-produced sex pheromone in the field. Our results suggest that in some plant bugs the male MSG esters have a natural communicative function as anti-sex pheromones, probably to repel other males from further mating attempts. From a practical standpoint, synthetic anti-sex pheromones may be useful for mating disruption of economically important plant bugs.

Sex pheromones in Diprionidae & other Symphyta (Hymenoptera).

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The most ancient group of Hymenoptera, Symphyta or sawflies, consists of around 8000 described species. The vast majority, are herbivorous and some species are important pests of agricultural crops or forest trees. Despite this, comparatively little is known about the chemical ecology within this group. I will present an overview of the sex pheromones identified so far in the different families, with emphasis on Diprionidae, the only well-studied group. In this family all active compounds discovered are saturated methyl-branched hydrocarbons with two to four chiral centres. The chiral configuration seems to be important for all species, whereas specificity regarding carbon chain-length or functional group (different esters) often is less distinct. The electrophysiologically and behaviourally active esters are released by the females together with their alcohol and carboxylic acid precursors. According to present knowledge, several sympatric species share the same sex pheromone, also with respect to stereoisomer, calling for more attention from chemical and evolutionary ecologists. In the other families the identified pheromones are different from those in Diprionidae, but there are too few sex pheromones identified for drawing any general conclusions.

Identification of volatile compounds from *Stenodema* (Heteroptera: Miridae) trigged by their natural predators.

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Gland secretions in Heteroptera contain different compounds spanning from sex pheromones to allomones. Little is known about the structure and function of released defence substances, especially in Miridae, and studies of emissions from living species are rare. In this study, *Stenodema* Laporte specimens were exposed to predators and the trigged chemical response was recorded in vivo using solid-phase microextraction (SPME). Sexual immature adults from *S. calcarata* Fallén, *S. holsata* Fabricius and *S. laevigata* Linnaeus were tested and predators like spiders and Nabidae were introduced to provoke defence reactions. Each species of *Stenodema* released three substances in high amounts due to the confrontation. These compounds were absorbed on SPME-fibers and identified using GC-MS.

Behavioural interactions between a slug pest & a carabid beetle predator.

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The co-evolution in the responses of predators and their prey is an exciting area of research which has practical benefits for the development of biological control. My work is focussed on how an important slug pest detects and responds to a carabid beetle predator and whether this can be related to the spatial dynamics of the predation interaction in the field. The avoidance response of the slug *Deroceras reticulatum* (Müller) to the carabid beetle *Pterostichus melanarius* (Illiger) was investigated under laboratory and semi-field conditions. Slug responses to the presence of both beetles and beetle odour were measured, and the chemical constituents of the odour which elicited the response were identified. Beetle odour had both a repellent and an antifeedant effect upon slugs and elicited anti-predator behaviour such as increased velocity and movement and a reduction in the turning rate of slugs. The findings were related back to data from a previous study that showed there to be a strong spatial relationship between the two species, indicative of density dependant predation on the slugs by the carabid beetle predator. The implications of this work for the efficacy of *P. melanarius* as a natural biological control agent of slugs will be discussed.

Functional diversification of the material properties of orb-weaving spider silk.

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Spider silks are exceptionally strong and can absorb more energy before breaking than high performance manmade fibers such as Nylon or Kevlar. The exceptional mechanical properties of spider silks are thought to have resulted in part from selection on the function of webs during prey capture, in particular upon aerial orb webs that must arrest fast-flying, high kinetic energy insect prey. However, orb-weaving spiders spin up to seven different types of silks, all of which are produced from distinct glands and are utilized in many different ways, from protecting eggs, to building retreats, to capture of prey. Here, we use the orb-weaving spider *Argiope argentata* to demonstrate that each of these silks, produced by distinct glands within a single spider, has a unique combination of material properties. Furthermore, these differences in mechanical performance reflect variation in the amino acid sequences of silk proteins. Thus, molecular evolution has played in important role in the evolution of the diverse toolkit of silks utilized by modern orb-weaving spiders.

Candidate pheromone receptors of the moth.

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The remarkable responsiveness of male moths to female released pheromones is based on the extremly sensitive and selective reaction of highly specialized sensory cells in the male antennae. These cells are supposed to be equipped with very distinct, male-specific receptors for pheromonal compounds; the nature of these receptors is still elusive. Using a combination of genomic sequence analysis and cDNA library screening of the tobacco budworm *Heliothis virescen***s led to the discovery of a divergent gene family encoding putative olfactory receptors. Comparing the encoded amino acid sequences of all candidate olfactory receptors emphasized their extreme diversity; however, a small family (identity > 40%) emerged, including the three receptor types HR14, HR15, HR16. RT-PCR-analysis revealed that all three subtypes were exclusively expressed in the antennae of male moths. More detailed** *in situ* **hybridization studies indicated that expression of these male-specific receptor types was confined to antennal cells located beneath sensillar hair structures which contain pheromone sensitive neurons. Moreover, two colour double** *in situ***-hybridization approaches in combination with laserscanning microscopy uncovered that each cell expressing one of these receptor type was surrounded by cells expressing pheromone binding proteins. These findings suggest that the novel receptor types may render neurons in male antennae responsive to pheromones.**

Influence of host footprints kairomone & past oviposition experience on the host location behaviour of *Trissolcus basalis.*

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During the host selection process insect parasitoid make flexible use of chemical information which can be directly or indirectly associated with the presence of their hosts. Wasp's response to a stimulus should be inversely related to its potential response and a change in wasp's response to stimuli can be the result of associative learning. In this view, influence of host indirect cues on the host location behaviour of the egg parasitoid *Trissolcus basalis* was investigated. *T. basalis* host selection behavior is mediated by chemical cues which are directly or indirectly associated with the presence of host egg mass of *Nezara viridula*. In particular, nymphs and adults footprints left on the substrate are known to play a role as indirect host-derived cues inducing an arrestment response in the parasitoid. We hypothesized that females which experienced the presence of host footprints kairomone without finding the host eggs should be less likely to respond to the same stimulus in the near future, while females that successfully located host egg masses would have shown reinforced responses. Results support our hypotheses. The significance of these findings for the manipulation of adult parasitoid behavior to enhance pest control and their implications for host selection by egg parasitoids are discussed.

Influence of direct mutual interference on patch time location in egg parasitoids.

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Direct mutual interference, i.e. the competition between parasitoid females foraging simultaneously for hosts, has regularly been considered to have a strong influence on the population dynamics of host-parasitoid systems. Furthermore, patch time allocation has also always been presented as an important behavioral component that should be deeply involved in the demographic features of host-parasitoid interactions. Based on a detailed analysis of within-patch interactions between *Trissolcus basalis* females, the present study aimed at understanding accurately how direct mutual interference can influence patch time allocation in this species. *T. basalis* attacks patches of its host, the egg masses of the Southern Green Stink bug *Nezara viridula*. In this species, competition between females occurs through strong fights when they are foraging simultaneously on the same patch. Such mutual interference could modify drastically the time invested by the females on the host patches. Behavioral interactions between the females were analyzed using a Cox's proportional hazards model. Results first indicated that the competitive interaction (through contacts and fights) between females strongly influences their patch residence time. Also, all the corresponding patch-leaving decision rules adopted by the females appeared to be under a significant genetic variation in the population studied. Such a genetic variability likely corresponds to an intra-population genetic variation in females' susceptibility to interference. The population dynamics and evolutionary consequences of these results are discussed.

Investigation of pheromones of coffee stemborers (Coleoptera: Cerambycidae).

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Stemborers (Coleoptera: Cerambycidae) are the most important insect pests of arabica coffee in Africa and Asia. In India and other countries of S.E. Asia, the main species is *Xylotrechus quaipes*. Adult males were previously shown to attract female beetles to traps in the field. Analysis of volatiles collected from virgin male beetles by gas chromatography (GC) linked to electroantennography (GC) showed three components accounting for over 90% of the volatiles, two of which elicited EAG responses from a female beetle. The major EAG-active component was identified as (*S*)-2-hydroxy-3-decanone (I) and the minor as (*S,S*)-2,3-dihydroxyoctane (II). The third component was 3-hydroxy-2-decanone (III). (*S*)-(I) was attractive to male *X. quaipes* in laboratory bioassays, but addition of (*S,RS*)-(III) at 10% of (I) reduced attractiveness. In field trials carried out in India with sticky, cross-vane traps, (*S*) and (*RS*)-(I) attracted male *X. quaipes* and addition of (*S,S*)-(III) at 10% of (I) reduced attractiveness. In Africa the main species is *Monochamus leuconotus*. Preliminary field studies have failed to show any attraction between the sexes, but collection and analysis of volatiles from male or female beetles has shown the males produce a compound not produced by the females and identification of this is in progress.

Chemical information in soil: tritrophic interactions between plants, insects & parasitic nematodes.

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Plants can protect themselves against insect herbivores by promoting the effectiveness of the herbivore's enemies. One way to do this is by releasing herbivore-induced volatiles that attract the herbivore's enemies. Most evidence for this phenomenon comes from studies on aboveground tritrophic interactions. We will discuss an example from underground tritrophic interactions: roots from a coniferous plant (*Thuja occidentalis*) release chemicals upon attack by weevil larvae (*Otiorhynchus sulcatus*). These chemicals attract parasitic nematodes (*Heterorhabditis megidis*) which are the natural enemies of the weevil larvae. We will discuss biological, chemical and exploitation aspects of this tritrophic interaction.

Collective foraging in an outbreaking forest pest.

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Gregarious behaviour is common among outbreaking forest insects. The forest tent caterpillar (*Malacosoma disstria*) is an early spring feeder that attacks North American hardwoods. Colonies exhibit complex social behaviour, using pheromone trails to move together between temporary bivouacs and feeding sites. Colonies alternate together between periods of quiescence and foraging, and mass movement is facilitated by chemical communication: moving caterpillars spin a fine thread of silk from their mouthparts and mark it with a highly stable non-volatile sterol pheromone. We show that the presence of trails significantly influences the behaviour of individual caterpillars, especially in the early larval stadia: trails increase both amount of locomotor activity and speed of travel. Larvae find novel food sources faster in an arena coated with trails than in a bare environment. This translates into a significant effect of trails on individual performance: caterpillars consume more food, grow larger and develop faster in the presence of trails. Although forest tent caterpillars do not recruit colony-mates to new food finds, trails nonetheless improve foraging by increasing individual mobility. These findings suggest that group-living is integral to the biology of the forest tent caterpillar and has shaped many aspects of its behaviour and development.

Finding the needle in a smelly haystack: prey location by carnivorous arthropods.

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Many natural enemies of herbivorous arthropods use herbivore-induced plant volatiles to locate their prey. These induced volatiles are sometimes referred to as 'SOS-signals' from plants and their composition is variable. The composition of a blend of herbivore-induced plant volatiles can vary with plant and herbivore species, and with other biotic and abiotic conditions. Carnivores have to cope with this variation to be able to locate their food. We investegated responses of a predatory mite to plant volatiles induced by its prey or by non-prey herbivores. The role of individual compounds from the volatile blend in the discrimination between different blends has been addressed, as well the importance of phenotypic plasticity in predator behaviour. Our data show that the predatory mite *P. persimilis* can locate their prey by using SOS-signals from the prey's host plant not only in setups that have no odour-background, but also when an odour-background consisting of other herbivore-infested plants is present. The induction of herbivore-induced plant volatiles is mediated by the phytohormone jasmonic acid. Application of this hormone can mimic the effect of spider mite infestation to some extent. Moreover, application of the phytohormone can increase the effect of spider mite infestations at low mite densities.

Interaction between olfactory & gustatory inputs in insect behavior.

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Behavior of insects is governed by chemical and physical signals impinging on specialized sensory organs. Volatile chemical signals are detected by the olfactory sense and result in upwind orientation by conspecifics receiving them. Detection of less volatile chemical signals by the gustatory sense results in behaviors such as copulation, biting and ingestion. However, seldom are these signals encountered in isolation; more often multiple chemical signals are detected simultaneously and the insect receiving them must process the information and appropriate a response. We investigated behavior of Colorado potato beetles challenged by simultaneous olfactory and gustatory stimuli. An open ambulatory Y-track olfactometer was used to measure insect preferences, number of turns made during orientation, and time to make a choice between stimuli. Olfactory stimuli were delivered to one-side of the device; gustatory stimuli were painted on the other side. Behavioral conflicts and preferences were discovered between stimulus pairs. For example, females took more time and made more turns when confronted with a volatile plant attractant and sucrose compared to the plant attractant and a male extract. We will examine behavioral hierarchies revealed by our studies and discuss how they may relate to reproduction and feeding.

Does the aggregation pheromone of *Riptortus clavatus* (Heteroptera: Alydidae) attract the *Piezodorus hybneri* (Heteroptera: Pentatomidae)?

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The male of *Riptortus clavatus* releases an aggregation pheromone which attracts conspecific adults and nymphs. We found that the synthetic aggregation pheromone (a mixture of three components) of *R. clavatus* (Heteroptera: Alydidae) attracted nymphs and both sexes of adults of *Piezodorus hybneri* (Heteroptera: Pentatomidae) in soybean fields. Among the three components of the synthetic pheromone, (*E*)-2-hexenyl (*E*)-2-hexenoate (EE) showed the attractiveness to *P. hybneri* and the activity was almost equal to the aggregation pheromone. The other two components lacked the attractiveness. Thus, EE was considered to be the component which showed the attractiveness to *P. hybneri*. Since we could not detect a peak of EE with the GC-MS analysis in the hexane extracts of *P. hybneri* and soybean plants, *P. hybneri* seems to utilize the component of the pheromone of *R. clavatus*, which has a similar host range, as a kairomone to search for food plants. However, real male bugs of *R. clavatus* did not attract the *P. hybneri*. Now we are trying to explain this contradiction.

Fatty acid levels & triacylglycerol biosynthesis in the sex pheromone gland of *Heliothis virescens* Fabricius.

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Most species of moths biosynthesize sex pheromone de novo in a specialized gland located between the 8th and 9th abdominal segments. As well as sex pheromone components, the gland contains large quantities of lipids, predominantly triacylglycerols. These triacylglycerols contain significant quantities of fatty acid precursors and other intermediates of the sex pheromone components. Despite the obvious role as a source of precursor acids for pheromone biosynthesis, the evidence in favor of this is equivocal; one particular problem is that insects using specific geometric isomer ratios for pheromone components tend to have very different ratios of the respective precursor acids. We believe that the glandular lipids have the role in pheromone biosynthesis of governing glandular fatty acid concentrations. When fatty concentrations are high (especially during high rate of pheromone biosynthesis), excess fatty acids are stored in the lipids; when concentrations low, fatty acids are mobilized from the lipids. This ensures the smooth production of pheromone during the sexually active period, which otherwise might be punctuated, for example, by high fatty acid concentrations inhibiting pheromone biosynthetic function. In this talk, we report the effect of glandular fatty acid levels on the biosynthesis of triacylglycerols in the pheromone gland of *H. virescens*.

Male mating behavior of the white-spotted longicorn beetle, *Anoplophora malasiaca* (Thomson) (Coleoptera: Cerambycidae) regulated by assorted female components.

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Male *A. malasiaca* dash towards and mount females, and bend the abdomen to copulate after contact with antennae or tarsi. Males make a similar mating attempt with glass dummies treated with the solvent extract of female elytra, indicating the presence of a female sex pheromone perceptible by physical contact. Chemical analysis of the pheromone components from female elytra revealed that at least three different groups of chemicals are involved: (1) long-chained alkanes, (2) ketones and (3) novel polar components. All three of these groups of chemicals are indispensable to evoke male mating behavior and work synergistically. Components within each group, however, complement each other. In addition to observation of the contact sex pheromone, we note the presence of volatile attractant produced by females in the near vicinity. Visual cues of females also appear to act synergistically with the volatiles in male mate location before direct contact by male antennae.

Control of sex pheromone precursor biosynthesis in adzuki bean borer *Ostrinia scapulalis.*

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The adzuki bean borer *Ostrinia scapulalis* is one of the eight *Ostrinia* species found in Japan, and is an important pest of adzuki bean plant and hop. The sex pheromone of *O. scapulalis* is a blend of (*E*)-11- and (*Z*)-11-tetradecenyl acetates. These components are biosynthesized from common saturated fatty acids through chain shortening, desaturation, reduction and acetylation. The key enzyme in this cascade is the Δ11 desaturase that introduces a double bond into myristic acid to form E11-/Z11-tetradecenoic acid (fatty acyl pheromone analog, FAPA). It is known that one day-old females of *O. scapulalis* do not show calling behavior. We found that these females had little FAPA in their pheromone gland while sexually mature 3 day-old females accumulated a substantial amount. As decapitated females did not synthesize FAPA, it appeared that a signal from cephalic organs is important for triggering FAPA biosynthesis. We further found that FAPA titer in the females maintained under a constant light condition is lower than that in the control females, suggesting that the light-off signal enhances FAPA synthesis. Since the expression level of Δ11 desaturase gene was constant regardless of FAPA titer, it was suggested that FAPA biosynthesis in *O. scapulalis* is under post-transcriptional regulation.

Relationship of visual & olfactory signal parameters in a food-deceptive flower mimicry system.

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Pollinators are attracted to flowers by their scent and visual display. While most flowers reward visits by providing pollen and/or nectar, some do not. Such mimicry relationships allow investigating the evolution of floral signals and their impact on pollinator behavior. We have reanalyzed a case of specialized food mimicry between *Orchis israelitica* and its model, the lily *Bellevalia flexuosa.* We characterized visual and olfactory signals of model, mimic, and of two phylogenetically related orchids. We mapped their visual signals to the perceptual space of honeybees and found a shift of the mimic's visual signals towards the model, confirming that visual mimicry is present. We analyzed flower odors using GC/MS. We related them to the perceptual space by testing them physiologically, using *in-vivo* optical recording. We found no evidence of olfactory mimicry. The results indicate that evolutionary processes altered *O. israelitica's* visual, but not olfactory, traits toward a higher similarity to its model. Apparently, odor mismatch does not prevent a bee from landing on a flower that has the expected visual display. The results therefore argue for the dominance of visual stimuli in short-distance flower choice. The orchid may still depend on long-distance olfactory attraction originating from neighboring model plants.

Pollinator responses to mimicry by sexually deceptive orchids.

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Deceptive orchids mimic insect signals to attract pollinators without providing a reward. Orchid flowers may resemble food, oviposition sites, or female insects. Insects that respond, by attempting to mate with or feed from orchid flowers for example, collect and distribute orchid pollinia. Deceived insects may incur costs, e.g. time and energy wasted travelling to and interacting with orchids, and loss of sperm during pseudocopulation. Such costs may encourage the evolution of behavioural strategies that allow insects to avoid deceptive orchids. In sexually deceptive orchids, male pollinators are initially attracted by scents identical to female insect sex pheromones. Short-range cues eliciting pollinator contact with the flower may include visual cues such as colour and shape, and structural or tactile features that resemble female insect bodies. Investigation of the relative importance of chemical, visual, and structural cues, and the interaction between, long- and short-range cues should be rewarding. The Australian orchid genus *Cryptostylis* is an excellent study subject because all five species attract the same male wasp pollinator despite obvious differences in flower shape and colour. Here I introduce this intriguing study system and present some initial observations of pollinator behaviour and interactions between pollinator learning and the dimensions of orchid patches.

Multimodal communication in the acoustic Lepidoptera: evolutionary pathways & biophysical constraints.

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Multimodal signaling, widespread among animal communications, may serve in conveying multiple messages, reaching different receiver categories, redundancy, and reliability. In Lepidoptera, sexual communication in various species involves male acoustic (ultrasonic) + chemical advertisement and female hearing + olfaction. These modalities operate during different stages of pair formation. Phylogenetic analysis indicates that olfaction is ancestral, with (male) chemical signaling and ultrasonic hearing (females, males) each evolving independently in multiple clades; hearing evolved as a response to predatory, echolocating bats. Finally, (male) acoustic signaling evolved in several clades possessing hearing. Dual use of the ultrasonic channel by acoustic Lepidoptera and bats indicates special problems for sexual communication in the former, as females must reliably distinguish male calls from bat echolocations. Moreover, they must distinguish these in flight and while perched, situations with exposure to different bats. Whereas multimodal communication may conceivably afford opportunities for adaptive response, studies of the pyralid moth *Achroia grisella* show that critical distinctions are made acoustically and via input from a third modality, proprioception: Females assess temporal characters of ultrasounds and respond appropriately in accordance with their assessment and situation, flying or in ground contact. This protocol may be particularly reliable, given the constraints of time and limitations of neural processing.

Patch leaving strategies & superparasitism: an asymmmetric generalized war of attrition.

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When several competitors deplete a patch, it can be advantageous for each of them to stay provided that others leave, whereas, on the other hand, staying longer decreases the expected payoff for everyone. This situation can be considered as a generalized war of attrition. Previous studies showed that when competitors interfere optimal patch leaving strategies become stochastic and the expected leaving time is much larger than predicted by the marginal value theorem. The possibility of superparasitism, as occurs e.g. in parasitoids, induces such interference. In addition, it gives several complications. First, the payoff of females that have left the patch is affected by the ovipositions of the remaining individuals. Second, differences in arrival time of females cause payoff-relevant asymmetries, since females that arrived early have parasitised more hosts in a patch at the moment superparasitism starts than those that arrived late. We show that this can be modelled as an asymmetric generalized war of attrition, and derive global characteristics of the ESS for simultaneous decisions when to start superparasitism and when to leave a patch

Reproductive strategies of cerambycid beetles & the role of semiochemicals.

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Reproductive strategies of adult cerambycid beetles, and the role of semiochemicals, are apparently associated with the condition of the host plant required by their larvae (whether hosts are alive, dying, or dead). For example, early studies have indicated that species whose larvae feed in living hosts are likely to use long-range pheromones, while those that feed in dying hosts typically rely on host plant volatiles to unite the sexes for mating. Nevertheless, more recent research has revealed evidence of long-range sex pheromones in dying-host species. There is also a growing body of evidence that mate recognition in species of both categories depends on contact chemoreception, and contact pheromones have been identified for several cerambycid species in recent years. Although the current state of cerambycid research documents considerable variation in reproductive strategy, the association with host plant condition provides structure for guiding future research.

Can female sex pheromones serve as a condition-dependent handicap trait?

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The handicap principle predicts that secondary sexual characters may act as honest signals of the individual quality if the trait bears a cost and if its expression is phenotypically condition-dependent. The cost of increasing the trait should be tolerable for individuals in good condition, but not for those in poor condition, thus, providing an honest signal of quality. Sex pheromones have long been recognized as important cues in sexual selection, however, whether sex pheromones comprise a handicap is unknown. We used the moth *Lobesia botrana* to study the role of sex pheromones as honest indicators of female quality because (a) females release a sex pheromone which attracts mate-seeking males from a distance, (b) there is evidence that females' calling behavior is affected by their size, (c) it was suggested that the female sex pheromone's characteristics are inherited and (d) males invest in the females more than sperm. We show that the male moths choose females as mates according to sex pheromone-related cues. We demonstrate that exposure to female species-specific synthetic sex pheromones bear a cost for the females and may therefore act as a condition-dependent trait and therefore we suggest that pheromones honestly advertise females' quality.

Mass trapping of olive moths.

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"A simple and inexpensive UV-trap was developed by the first author from materials generally available around the olive farm, to capture the main lepidopterous pests of olive trees. The trap consisted of a sticky-collecting device, two ultraviolet fluorescent tubes and tilted mirrors to increase light intensity by interference of reflected rays and spreading light reflection over a wider area. The trap was compared with delta-traps (baited with (Z)-Z-tetradecenol) and funnel traps (baited with (E)-11-hexadecenol and (E)-11-hexadecenyl-acetate) for monitoring the olive moth, *Prays oleae* and jasmine moth, *Palpita unionalis*, respectively. Sex pheromone catches (only males) were persistently lower in all collections of 2 successive olive seasons of 2002 & 2003 than those of UV-traps. *P. oleae* catches (males & females) in the UV-trap significantly ($P<0.01$) exceeded *P. unionalis* moths. The obtained results indicated that the UV-trap may represent an effective tool for mass-trapping of *P. oleae* and monitoring tool for *P. unionalis* and other species of moths in the area. The trap was so effective in trapping *P. oleae* even when adult population was low compared with sex pheromone trap. Some of non-target species may be protected from damage by placing a screen of proper sized mesh over the entrance of the trap. The UV-trap saved the fruit yield of a large olive farm without using any insecticides throughout the study period.
"

New approaches to find key compounds in tritrophic interactions – exploiting genetic variation & *in situ* modification of volatile profiles.

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Odour blends that are released by plants after herbivore attack can act as indirect defense by attracting natural enemies of the herbivore. These blends consist of mixtures of volatile organic compounds (VOCs), but, as yet, for many tritrophic systems it remains unknown, which VOCs of such complex blends are the most attractive ones. One way of studying the importance of individual VOCs is to test the attractiveness of odour blends differing in their pattern of compounds. This approach was used to test the attraction of naive and experienced females of the parasitoid wasp *Cotesia marginiventris* to volatile blends of maize infested with *Spodoptera littoralis* larvae. We used olfactometers in which female wasps were offered choices among odour blends of different chemical composition. The different blends were obtained by either using different maize inbred lines or by installing selectively adsorbing filters between the odour source and the olfactometer arms. The data were analysed with multivariate statistical techniques and revealed the relative importance of some VOCs that are often underestimated because of their small quantities.

How parasitoids read the news: information use in a patchy world.

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Patch leaving decisions in insect parasitoids have mainly been studied as a function of host density and host encounters in single-patch experiments. From such experiments we know that parasitoids respond to host kairomones and encounters with unparasitized hosts. Yet, parasitoids often encounter unparasitized as well as parasitized hosts. Response to encounters with parasitized hosts has been debated, but experimental evidence has been equivocal. Moreover, how information on a given patch and patch encounter rate influence the patch time allocation on subsequent patches has largely remained unanswered. Using *drosophila* parasitoids, we investigated behavioural responses of wasps to information cues for estimating patch quality and habitat quality. In patch-quality experiments we tested responses towards staged encounters with unparasitized vs. parasitized hosts. In habitat-quality experiments we tested whether odour from neighbouring patches affect the patch-time, and the searching behaviour when leaving the patch. Further, we studied how encounters with good and poor quality hosts and their density influence the time allocation on subsequent patches. Finally, we tested how parasitoids respond to variation in patch encounter rates in their patch time allocation and patch exploitation level. We discuss our results with respect of the reliability of such cues for future foraging success.

Floral scent mediating the foraging behaviour of some papilionid & danaid butterflies.

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In addition to visual stimuli, olfactory cues also play a significant role in the foraging behaviour of anthophilous butterflies. I investigated floral volatiles of *Citrus* flowers (white in colour) that stimulate flower-visiting by a papilionid butterfly, *Papilio protenor*, which shows innate colour preference for red, orange, and blue. To determine active compounds, odour components were first screened individually to assess their ability to evoke proboscis extension reflex (PER), and the actual attractancy of potential components was confirmed by free-flight experiments. Of 16 compounds identified from the flower, methyl anthranilate, 2-phenylethanol, and phenylacetonitrile were found most attractive to the butterfly, indicating that compounds with an aromatic ring serve as important olfactory cues in their flower-visiting. In Japan, flowers of *Eupatorium chinense* have been observed to strongly attract danaid butterflies in the genera *Parantica, Ideopsis, Euploea,* and *Salatura*. Although several volatile compounds detected from an isopentane extract of the flower showed weak to moderate PER-activities, none of them exhibited strong attractiveness to the danaids. However, floral nectar of the plant was found to contain a considerable amount of pyrrolizidine alkaloids. The major compound, identified as intermedine, strongly attracted some danaid species, suggesting that certain pyrrolizidine alkaloids *per se* function as attractants.

Molecular cloning of a putative PBAN receptor from pheromone glands of the silkmoth, *Bombyx mori*.

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Despite recent successes in elucidating the molecular components of sex pheromone biosynthetic pathways within the Lepidoptera, identification of the gatekeeper of the pathway, the PBAN receptor, has proven to be elusive. Given the sequence similarity between the active C-terminal regions of PBAN and neuromedin U, it was hypothesized that their respective receptors might also be similar in structure. Consequently, a homology-based molecular approach utilizing primers constructed from the conserved regions of insect neuromedin U receptor homologues was used to amplify a full-length 2777-nt clone from a *Bombyx mori* pheromone gland cDNA library. Conceptual translation of the ORF indicated that it encoded a 45.8 kDa 7 transmembrane protein with 82% identity to a GPCR that has been recently cloned from pheromone glands of *Helicoverpa zea*. Interestingly, the sequence identity decreases considerably within the amino terminus and the *Bombyx* receptor contains a 67 amino acid C-terminal extension. Tissue distribution analyses reveal that the receptor is specific to the pheromone gland where it undergoes significant up-regulation in the day preceding eclosion. In addition, when expressed in Sf9 cells the receptor not only exhibits specific binding to PBAN but also mobilizes calcium in a dose-dependent response.

Reception of floral odours by *Helicoverpa armigera* antennae.

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The antennal receptor cell complex of *Helicoverpa armigera*, that is used in detecting a range of floral and green leaf odours, was investigated with electrophysiological and behavioural methods. By using mixtures of odours at saturating levels, the electroantennogram method can produce useful information about the classes of receptor cells used in detecting plant odours. This methodology provides information that both differs from and augments that gained by single neuron recordings, and as such allows the testing of hypotheses related to receptor types. A range of volatiles was tested including those principally associated with flowering parts or leaves as well as volatiles common to both. The two enantiomers of the chemical μ-pinene were also investigated and results suggest a common receptor neuron type for these enantiomers. Behavioural experiments showed that conditioned learning of odours can influence the decisions of the insect.

Significance of male's licking female dorsum in matings of Cerambycidae (Coleoptera).

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In matings of, if not all, some coleopteran families including Cerambycidae, it has been often recognized that male, on female's back, lick her dorsum with his mouth parts. However, there has been only a very limited number of literature on this behavioral component. We therefore investigated the significance of each of the body parts in cerambycid mating sequence, focusing on males' dorsum-licking behavior. Adults of diurnal cerambycid species, *Dinoptera minuta* (Gebler) (Lepturinae) and *Chlorophorus quinquefasciatus* (Cerambycinae), were collected in the field, and each of the test animals was treated by mutilating antennae with scissors (male, female) or coating its compound eyes (male, female) or pronotum plus scutellum (female) with water-based white correcting fluid. A treated individual and an untreated partner were paired and their mating sequence followed. Untreated pairs initiated mounting directly upon male antenna's touching female's body or antenna. Antenna-mutilation of male caused severe hinance of mating. Blind males did not mount or copulate. Coating pronotum/scutellum-region of the female also caused copulation hinance. The last result suggests that female beetle possibly possesses in her pronotum/scutellum region a source of pheromone toward the male and/or a sensory organ admitting male's appeasement by his mouth parts while in copula.

Using synthetic herbivore-induced plant volatiles to enhance conservation biological control in crop ecosystems: field experiments in hops & grapes.

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The potential of using synthetic herbivore-induced plant volatiles to enhance conservation biological control of insects and mites in crop ecosystems is being researched in hops and grapes in Washington State. Results to date indicate that a number of natural enemy species in the families, Chrysopidae, Hemerobiidae, Anthocoridae, Geocoridae, Miridae, Coccinellidae, Syrphidae, Braconidae, Empididae and Mymaridae, are attracted to sticky traps baited with aqueous methyl salicylate (MeSA), hexenyl acetate, farnesene or octyl aldehyde. Further, hop yards and grape blocks baited with slow release sachet dispensers of MeSA appeared to recruit larger populations of some insect predators (e.g. *Stethorus punctum picipes, Orius tristicolor, Chrysopa nigricornis*) than unbaited areas, resulting in some instances, in improved biological control of spider mites. The use of synthetic plant-signalling compounds as 'Herbivore-Induced Plant Protection Odors' (HIPPOs) may provide a novel yet practical option for improving the efficacy and reliability of conservation biological control in a variety of agricultural ecosystems.

Mating behaviour of the New Zealand native leafroller *Cnephasia jactatana* Walker (Lepidoptera: Tortricidae).

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Cnephasia jactatana Walker is a pest of kiwifruit in New Zealand. The objective of this study was to provide information on its mating behaviour for the development of pest management measures. Sexual maturation period was 3 d for females and 2 d for males. More than 50% of eggs were laid within 3 d after mating. Female reproductive fitness was reduced by > 85% if mating was delayed for 4 d. Males actively approached and courted females before mating occurred and females appeared to be less active. Males used their antennae, labial palps and wings to touch females when courting. Virgin males performed more mating attempts than mated males when competing for females. Sexual selection in *C. jactatana* did not support Darwin's sexual selection theory where females are more selective and males suffer the sexual selection effects. In this species, males appeared to be more selective than females. Both sexes preferred virgin to mated mates for mating. Males preferred young to old females for mating while females did not discriminate the male age when choosing a mate. Male body weight did not confer any mating advantage. However, females preferred males with longer antennae for mating. Light- and average-weight males presented a size assortive-selection while heavy males did not show any preference.

Regulation of pheromone biosynthesis in moths: PBAN & a G-protein coupled receptor.

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Pheromone biosynthesis in most moths is regulated by PBAN (pheromone biosynthesis activating neuropeptide). This hormone is produced by the subesophageal ganglion and can be released into the hemolymph to stimulate biosynthesis in pheromone glands. PBAN acts directly on pheromone glands using cAMP and calcium as second messengers. Biosynthesis is activated by stimulating acetyl-CoA carboxylase or a reductase. Pharmacological evidence indicated the presence of a G-protein coupled receptor. To identify the receptor we utilized the finding that *Drosophila* contains PBAN-like peptides and thus a receptor for the peptides. Three *Drosophila* receptors had sequence homology to neuromedin U receptors from vertebrates. Neuromedin U will stimulate pheromone biosynthesis in *Helicoverpa zea* and *H. armigera* indicating cross-reactivity of the peptide and functional homology of the receptors. Based on sequence homology to the receptors from *Drosophila,* the open reading frame of a gene was isolated from mRNA obtained from pheromone glands of *H. zea*. The PBAN-receptor was cloned and expressed in Sf9 cells and using a calcium mobilization assay responded to PBAN in a dose-dependent manner. Additional peptides produced by the PBAN family were also tested. This represents the first identification of a receptor for the PBAN family of peptides with a known function in female moths.

Herbivore-induced volatiles alter plant resistance.

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In response to damage, several plants have been found to emit volatiles that make neighboring plants more resistant to herbivory. When sagebrush is damaged in the field, airborne volatiles cause neighboring conspecifics to change chemically and suffer less damage by naturally occurring herbivores. Emissions of methyl jasmonate increase in the headspace of damaged sagebrush, making it a leading candidate as a volatile cue mediating increased resistance. Since many plant species respond to methyl jasmonate, it seemed plausible that various neighbors might induce resistance when growing near damaged sagebrush. Damage to sagebrush in the field caused neighboring wild tobacco but not other naturally occurring plant species to become more resistant. Preliminary laboratory experiments suggest that methyl jasmonate is sufficient, but not necessary, as an airborne cue.

Effective pheromone lures for monitoring the rice leaffolder moth, *Cnaphalocrocis medinalis* **(Lepidoptera: Crambidae).**

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Several factors affecting the attractiveness of traps baited with synthetic sex pheromone components of *Cnaphalocrocis medinalis* were examined in the laboratory and field. In the laboratory, among the four pheromone components, Z11-18:Ald, Z13-18:Ald, Z11-18:OH and Z13-18:OH, the major component, Z13-18:Ald, was shown to be an essential component for attraction and Z11-18:Ald was shown to be a potent synergist for the major component. The alcohols also showed an apparent synergistic effect in the laboratory bioassay when their mixture was combined with an aldehyde mixture. The addition of the geometric isomers of the four components at levels greater than 5% of the purified 4-component blend showed significant inhibitory effects on the pheromonal activity in the laboratory. In the field, no synergistic effect of the alcohols on the aldehydes was shown. A range of loadings of the pheromone rubber septa (0.01–3.42 mg) was tested and all remained attractive for at least one month with optimum attractiveness on 0.86 ㎎/septum. No significant effects were obtained when the ratio of Z11-18:Ald to Z13-18:Ald was changed. Rubber septa containing 0.86 mg of the four components (*E*-isomers 5%) at the natural ratio are being effectively used for monitoring.

Experimental evolution of improved learning ability in *Drosophila.*

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While fitness benefits of animal learning are well understood, other questions concerning the evolution of learning remain open. How easily can improved learning ability evolve? Does evolution act on "general intelligence", or rather on learning abilities specialized to particular tasks and situations? What are the costs of learning ability? To address these questions we have exposed replicated populations of *Drosophila melanogaster* to long-term selection for improved olfactory-gustatory learning in the context of oviposition substrate choice. After over 20 generations of selection the experimental populations evolved markedly improved learning ability. The improved learning ability generalized to other tasks, which involved different stimuli, reward rather than aversion learning, and classic rather than operant conditioning. The selected populations also showed better learning ability as larvae. This indicates that the response to selection was based on evolutionary changes in general aspects of the learning and memory, involved in many forms of learning. Furthermore, compared to control populations, larvae of the selected populations showed lower competitive ability. Additional data excluded inbreeding as a potential cause of reduced competitive ability. Therefore we believe that the lower competitive ability of the experimental populations indicates a genetic cost of learning ability.

Patch leaving decisions of *Cotesia rubecula*: a synthesis.

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Cotesia rubecula (Hymenoptera: Braconidae) is a solitary endoparasitoid of larval *Pieris rapae* (Lepidoptera: Pieridae). Adult P. *rapae* lay single eggs on a range of *Brassica* species (Brassicaceae) and other plants, and their larvae develop as individuals on host plants. *C. rubecula* parasitises first through fourth instars of *P. rapae.* A series of laboratory and field experiments has examined patch-leaving decisions of Australian *C. rubecula.* A range of factors has been shown to influence patch-leaving tendency of this parasitoid including host stage and density, the amount of plant damage by the host, recent oviposition experience of the wasp and the wasp's feeding status. Decisions are also influenced by the spatial distribution of host plants. This paper will develop a synthesis of experimental results and aims to identify the selective factors that have shaped the foraging behaviour of *C. rubecula.*

The infrared organ of the Australian "little ash beetle" ***Acanthocnemus nigricans*****: morphology of the specialised sensilla.**

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The Australian beetle *Acanthocnemus nigricans* (Coleoptera, Acanthocnemidae) approaches forest fires. The reason for this pyrophilous behaviour might be that larval development depends on burned wood. *A. nigricans* has a pair of infrared organs on the ventrolateral sides of its prothorax. Each organ consists of a cuticular "sensory disc"with a diameter of about 150µm. About 70 small cuticular sensilla were found mainly on the outer surface of the disc. The cone shaped cuticular peg of a sensillum is about 2µm in diameter. It has no pores and is surrounded by a cuticular wall. The single sensory cell of the sensillum shows some similarities to the "bauplan" of a mechanoreceptive sensory cell of a trichoid sensillum. However, three specialisations can be found: (i) the denitic outer segment is enveloped by a pronounced electron-dense structure, which most likely represents the hypertrophied denitic sheath. (ii) The membranes of the denitic inner segment and the soma show distinct infoldings which contain a high amount of mitochondria. (iii) No tubular body can be found.
As the cuticular disc responds to infrared radiation, it is likely that this specialised new type of insect sensillum may act as a thermoreceptor. Further examinations will have to test this hypothesis.

Pheromone response & mating activity in the consperse stink bug, *Euschistus conspersus.*

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The consperse stink bug, *Euschistus conspersus* Uhler, has emerged as an important pest of pome fruit production in Washington state. My research has focused upon quantifying the behavioural response of this insect to its aggregation pheromone, methyl (2*E*,4*Z*)-decadienoate in an attempt to develop pheromone-based monitoring or management systems. I describe research investigating the pheromone-mediated aggregation behaviour in *E. conspersus* in the field and describe the subsequent development of a detailed diel activity profile for this insect. *E. conspersus* aggregations peaked at dusk, with mating occurring primarily at night. Sex ratios were equal in aggregations and both sexes were found to be polygamous, mating on successive nights but with different partners. Laboratory experiments were performed to investigate possible factors mitigating mate choice in this species. The significance of these findings with respect to the possible function of aggregation in this species is discussed, as well as the implications for management.

Molecular basis of pheromone binding, release, & inactivation.

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Insects perceive the world through small molecules - pheromones and other semiochemicals. Their odor-oriented navigation relies heavily on the ability of the olfactory system to selectively detect minute amounts of pheromones and reset the detectors in millisecond scale (by inactivating the chemical signals). Pheromone molecules enter the sensillar lymph through pore tubules in the sensillar wall, are solubilized upon being encapsulated by odorant-binding proteins (OBPs), and transported to the olfactory receptors (ORs). Upon interaction with negatively-charged sites at the denitic membrane, the OBP-ligand complex undergoes a conformational change that leads to the ejection of pheromone to activate the pheromone receptors. While bound to OBPs, pheromone molecules are protected from odorant-degrading enzymes (ODEs). In this presentation, we will (1) briefly discuss the molecular mechanisms of pheromone binding and release by focusing on the *Bombyx mori* system, i.e., how bombykol enters and leaves the binding cavity in BmPBP, (2) report the first odorant-degrading enzymes isolated from insect antennae to date, and (3) discuss the possible mechanism of enzymatic inactivation of pheromones.

Induction of preference for oviposition repellents originated from non-host plants by a specialist herbivore.

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Repellents derived from non-host plants have been assumed to have a role to play in the behavioural manipulation of crop insect pests. However, behavioural responses to such repellents by insects have been rarely observed either in the laboratory or field. In laboratory and greenhouse experiments, we observed the responses of diamondback moth, *Plutella xylostella*, a special herbivore of crucifer plants to oviposition repellents derived from a non-host plant, *Chrysanthemum morifolium.* When provided with a choice between untreated Chinese cabbage and Chinese cabbage sprayed with an extract of *C. morifolium* in one arena, inexperienced females deposited 9-fold more eggs on the former than on the latter plant, demonstrating that the plant extract had substantial repellent effects on this insect. However, when the test adults were offered prior experiences of the plant extract and then exposed to the dual-choice of plants for oviposition, they laid significantly more eggs on the plant sprayed with the *C. morifolium* extract than that on the untreated plant. Further observations with neem-based materials also showed induction preference to these repellents by *P. xylostella.* Adult learning thus can render repellents derived from non-host plants to become ineffective or even attractive to this insect.

Non-host antifeedants in hylobius pine weevils: from lab to field applications.

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We searched for antifeedant activity in non-host woody plants to find new compounds for seedling protection of Scots pine (*Pinus sylvestris*) and Norway spruce (*Picea abies*) against pine weevil feeding. In total, 38 species from 25 families were compared. The weevils removed less of both outer and inner bark in *Ilex*, *Evonymus*, *Populus*, *Syringa*, *Taxus*, *Tilia*, *Viburnum*, *Lonicera*, and *Sorbus* than on *Pinus*. In some plants, *Juglans*, *Fraxinus*, *Sambucus*, *Aesculus*, *Quercus*, *Corylus*, *Fagus*, *Salix*, *Alnus*, & *Acer*, the outer bark was removed by the insects but had an inner bark with antifeedant qualities. Bark of Linden, *Tilia cordata*, showed the best antifeedant qualities and was extracted and tested for antifeedant activity. The active extract was fractioned on silica gel. TLC and GC was used to trace chemical similarities between the fractions and resulted in merging the number of fractions down. The micro assay gave positive antifeedant activity in two adjacent fractions. These fractions were analyzed and nonanoic acid was found in both of them. Synthetic octanoic, nonanoic, and decanoic acid tested in the micro feeding assay had strong antifeedant activities against both sexes.

Characterization of genes encoding enzymes involved in bombykol biosynthesis.

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In the biosynthesis of class I-type Lepidopteran pheromone components, the key enzyme required to produce the oxygenated functional groups is pgFAR. This enzyme converts fatty-acyl pheromone precursors to their corresponding alcohols, which can then be, depending on the moth species, further modified via acetylation or oxidation reactions. In experiments designed to characterize the pgFAR in *B. mori*, we have isolated a cDNA clone encoding a protein homologous to a FAR from the desert shrub, *Simmondsia chinensis*, commonly known as jojoba. The deduced amino acid sequence of this clone predicts a 460 amino acid protein with a consensus NAD(P)H binding motif within the amino terminus. Northern blot analyses indicated that the 2-kb transcripts of this gene are specifically expressed in the pheromone gland at one day prior to adult eclosion. Functional expression of this gene in the yeast *Saccharomyces cerevisiae* not only confirmed the long-chain FAR activity but also indicated a distinct substrate specificity. Finally, the transformed yeast cells evoked typical mating behavior in male moths when cultured with the pheromone precursor fatty acid. We will also refer to a bifunctional desaturase utilized in the production of the conjugated diene system from which bombykol biosynthesis is dependent on.

"Come on baby light a fire": the reproductive biology of *Actebia fennica.*

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Black army cutworm out breaks are associate recently burned areas, which are unpredictable in time and space. Our wind tunnel experiments showed that while females exhibit oriented, upwind flight in smoke plumes males do not. We also examined the effect of smoke volatiles the temporal changes in juvenile hormone biosynthesis and oocyte development, as well as on the age at which females first exhibited the behaviour associated with the emission of the sex pheromone. All data support the hypothesis that volatiles from burnt vegetation serve as " kairomones", accelerating sexual maturation in females. However, mated females, held in the presence of volatiles that stimulated sexual maturation, actually laid fewer eggs. An ecological hand-waving scenario will be proposed to explain the results obtained to date. We will also discuss how the recent identification of different smoke compounds detected by females might subsequently be used in management programmes for this pest species.

On the kairomonal attraction of adults & nymphs of *Elatophilus* spp. (Heteroptea: Anthocoridae) to sex pheromones of *Matsucoccus* spp. (Hemiptera: Matsucoccidae).

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While kairomonal response of insect predators to the sex pheromone of the insect prey has been rarely documented, our recent studies proved consistently that the female sex pheromones of *Matsucoccus josephi* , *M. feytaudi* and *M. matsumurae* are powerful long range attractants for *Elatophilus* spp. Examining the attraction to the pheromone and analogs of seven *Elatophilus* spp. revealed a clear species specific attraction displaying by these predators. Similarly to their conspecific adults the nymphs of *E. hebraicus, E. crassicornis* and *E. nigricornis* are also lured by the specific sex pheromone. It seems that sex pheromones of *Matsucoccus* serve *Elatophilus* spp. as aggregation kairomones and as sexual stimulants. *Elatophilus* spp. display courtship behavior and mate frequently in arenas baited with the pheromone. When the predator population is high, most of the females that congregated around a pheromone source were already mated but displaying none or few visible eggs in the ovaries. We suggest that the typical group movement displayed by the nymphs is determined by their attraction to the pheromone. This behavior may increase survival through "a better feed tracing" and "safety in numbers". Female first mating probably occur soon after molting within their own group before being lured to a pheromone source.

Odor perception & odor learning in honeybees.

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Honeybees discriminate a large range of odors, and learn odors as signals for food. We have used optical imaging and intracellular recording techniques to examine the neural substrates of odor coding and memory formation in the antennal lobe (AL) and the mushroom bodies (MB). Odors elicit combinatorial patterns of activity in the glomeruli of the AL. Imaging of the input region of the mushroom bodies reveals that odors are represented in specific patterns of microglomerular activation. A comparison of the dynamics of the odor responses shows that mushroom body intrinsic neurons code odors in a sparse way with a small number of spikes per stimulus and a small number on activated Kenyon cells. Odor learning leads to an enhancement of the synaptic transmission at the input synapses to the Kenyon cells that respond to the trained odor (CS+), and a reduction of synaptic efficiency to the non trained odor (CS-). A model will be presented that captures the results of our electro- and optophysiological recordings, and assigns particular functions to the mushroom body in the bee brain.

Male-produced aggregation pheromone of *Neoclytus acuminatus acuminatus*.

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The red-headed ash borer, *Neoclytus acuminatus acuminatus* (F.) (Coleoptera: Cerambycidae) is endemic to North America, where it infests a variety of hardwood trees. It also has been accidentally introduced into Europe and possibly New Zealand. In the field, we observed males performing a stereotyped calling behavior which provided the first indication of pheromone production by male *N. a. acuminatus*. This behavior has not been reported for any other cerambycid species. Insects of both sexes were attracted to caged males but not to females. We report the identification and field testing of a male-produced aggregation pheromone for this species. During field trials, females of another cerambycid species, *Curius dentatus* Newman, were attracted to traps baited with some of the test compounds, suggesting that one of more of the test compounds were also part of the sex pheromone of this relatively unknown species as well. These may be the first examples of cerambycid pheromones with an activity range of tens to a huned meters or more.

Pheromonal & vibrational signals of stink bugs in the genus ***Chlorochroa*****.**

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Stink bugs in the genus *Chlorochroa*, similar to other phytophagous stink bugs, use a combination of pheromonal and substrate-borne vibrational signals in their reproductive behaviors. The pheromones are male-produced, and probably act as attractants over longer distances, whereas vibrational signals (songs) are produced by both sexes once the bugs are on the same plant. The vibrational signals are sex- and species specific, with males and females producing a repertoire of calling songs as orientation cues, and courtship songs once the bugs are close together or in contact. The pheromonal and vibrational signals will be compared and contrasted for three sympatric *Chlorochroa* species, and the roles of these signals in intraspecific communication will be discussed.

Fruitspotting bugs.

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The fruitspotting bugs, *Amblypelta nitida*, and the closely-related *Amblypelta lutescens*, are serious pests of tropical and sub-tropical fruit and nut crops in Queensland and northern New South Wales. Current control relies on the frequent application of insecticides, with recent investigations focussing on promising soft-option strategies such as restricting insecticide application to fruitspotting bug hotspots, and the use of SurroundÒ to deter feeding. Behaviour-modification approaches are a legitimate component of clean-green management strategies, and to this end, investigations of the insects' semiochemistry were commenced 14 years ago. Despite early breakthroughs in the chemistry studies, subsequent progress has been sporadic. The major component of male-derived volatiles of *A. nitida* was shown to be a mono-epoxide of (E)-b-ocimene, having the trivial name, myroxyde; and of *A. lutescens*, (-)-(3R)-(E)-nerolidol. The absolute configuration of the former compound has been defined through the development of efficient synthetic strategies and the application of SPME sampling technology. Recent gas chromatography-electroantennography studies have confirmed myroxide, along with the minor compounds, nonanal and decanal, as stimulants of antennal response in *A. nitida*. The attractiveness of blends of racemic myroxyde and the aldehydes, using a new clear-plastic football-shaped trap, is being investigated.

Exposure to allelobiotic substances has a physiological effect on cereal plants, evidenced by significant changes in the pattern of biomass allocation and changes in leaf temperature.

Isolation & characterisation of a putative aggregation pheromone component for the striped cucumber beetle, *Acalymma vittatum* (F) (Coleoptera: Chrysomelidae).

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Male-produced aggregation pheromones have previously been identified from a limited number of Chrysomelids; the hydroxy-ketones (*S*)-3,7-dimethyl-2-oxo-6-octene-1,3-diol from the Colorado potato beetle, *Leptinotarsa decemlineata* (Say) [1], and (*E*)-8-hydroxy-6-methyl-6-octen-3-one from the cereal leaf beetle, *Oulema melanopus* L. [2]. Himachalene and cadinene sesquiterpenes from *Phyllotreta cruciferae* and three *Apthona* spp. of flea beetles were electrophysiologically active [3]. Field trapping studies showed that feeding male *A. vittatum* attracted significantly more congeners than feeding females, or undamaged cucumber plants, indicating that it may also use an aggregation pheromone [4]. GC-EAD studies were carried out using airborne collections from feeding beetles, or SPME collections, and male or female antennae. One component of male collections consistently gave a strong EAD response, and was absent from collections from female beetles. This putative pheromone was initially characterised by GC-MS and micro-reactions. More than 3000 beetle-days of collection on Porapak resulted in sufficient compound for extensive 1- and 2-D NMR spectroscopic studies, and FTIR. These allowed complete structural characterisation of the pheromone. The only closely related compounds are antibiotics of microbial origin, which were compared with the pheromone by NMR. Currently, efforts to synthesize the pheromone, and test for behavioural activity are in progress.

Sex pheromone of a carpenter moth, *Cossus insularis* **(Staudinger).**

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Cossus insularis (Staudinger) (Cossidae: Lepidoptera) is a woodborer found in Honshu, Kyushu and Tsushima Island of Japan. The larvae excavate their galleries in a group on twigs or trunks of the broad-leaved trees such as willow trees, pear trees, chestnut trees, and cause damage and often mortality of the trees. To utilize sex pheromone for monitoring or controlling the borers, sex pheromone of *C. insularis* was identified. Combined gas chromatography-electroantennographic detection (GC-EAD) analysis of airborne volatiles collected from unmated female moths by a solid-phase microextraction (SPME) showed that two volatile compounds elicited significant electroantennographic responses from the antennae of male moths. These two volatile compounds collected from female moths were identified as (*E*)-3-tetradecenyl acetate and (*Z*)-3-tetradecenyl acetate by mass spectral and retention index comparisons with synthetic standards. In field bioassays, sticky traps baited with a lure of a 4:1 blend of synthetic (*E*)-3-tetradecenyl acetate and (*Z*)-3-tetradecenyl acetate captured significantly large number of male moths, while unbaited traps captured no male moths. These results indicate that sex pheromone of *C. insularis* is a mixture of (*E*)-3-tetradecenyl acetate and (*Z*)-3-tetradecenyl acetate.

Plant/plant communication-herbivores & their natural enemies: experiments with spring-sown barley.

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Experiments with a barley crop and weeds have shown that effects of chemical communication between plants can be traced in a tritrophic perspective as follows:
Exposing cereal plants to allelobiotic volatiles from neighbouring plants and root exudates from aggressive weeds decreases the acceptability of the exposed plants for aphids. This happens in interactions between different cultivars of barley (*intra-species*), and between cereals and a number of aggressive weeds (*inter-species*).
Exposure to allelobiosis from weeds causes barley plants to become more attractive to aphid natural enemies, a ladybird *Coccinella septempunctata.*
The results suggest that plant responses to allelobiosis is a preparation for coexistence with other plants and this is a basis for follow up effects in higher trophic levels. The positive effect on searching behaviour of polyphagous predators may increase the ecological success of "listening" plants in a mixed plant stand and, also have negative effects on herbivore plant acceptance.

Artificial substrate bioassay for testing oviposition of the southern green stink bug, *Nezara viridula* (L.) conditioned by soybean plant chemical extracts.

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The southern green stink bug, *Nezara viridula* (L.) (Heteroptera: Pentatomidae) is effectively controlled with egg parasitoids, and egg production is an important component of this strategy. Therefore, a laboratory bioassay was developed for testing its oviposition preference toward chemicals extracted from soybean pods and leaves. An artificial substrate (cheesecloth) was stretched over a wooden ring (embroidery hoops), treated with plant extracts or chromatographic fractions, and then exposed to adult stink bugs to assess oviposition preference. Freeze-ied soybean leaves and pods were extracted with hexane, then with methanol, and finally with water. The organic solvent extracts were fractionated by liquid chromatography, on a C_{18} reverse phase column, on a normal phase silica column, and by thin layer chromatography (TLC) on silica gel. The methanol extract of pods showed the greatest bioassay activity. The most active reverse phase fraction derived from this extract was eluted with 20% methanol in water. After subjecting this fraction to chromatography, the greatest activity occurred in the fraction eluted with 60% methanol in methylene chloride. TLC gave no single fraction with demonstrated activity, but the recombined fractions were again active, indicating that multiple components elicit oviposition. Hairs (long and short) on the female genitalia might be involved in chemical and/or mechanical recognition of oviposition stimulants.

The chemistry of sexual deception in Australian orchids.

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Pollination by sexual deception is among the most intriguing of orchid pollination syndromes. Odors are well established as the primary stimuli for sexually attracting the male insect pollinators in these orchids, but we have only begun to discover how many, and which compounds are involved. Using gas chromatography with electroantennographic detection (GC-EAD), GC-mass spectrometry (GC-MS) and GC-IR-spectrometry we have determined that a single active compound, Chiloglottone, produced by both females of the wasp *Neozeloboria cryptoides* and flowers of the orchid *Chiloglottis trapeziformis* is active in antennal neurons of the pollinators, males of *N. cryptoides*. This compound, tested as synthetic copy, proved to be equally attractive to natural extracts of flowers and wasps. We are now determining the chemical identity of sexual attractants in other *Chiloglottis* orchids. GC-EAD and GC-MS analysis indicates only 1 or 2 active compounds are involved and these compounds are related to Chiloglottone. Pollinator specificity and associated reproductive isolation is therefore likely under simple genetic control in these orchids. Thus rapid sympatric speciation is possible. We will present an update of our multifaceted research program that is exploring this hypothesis of sympatric speciation.

Biochemistry & evolution of floral scent.

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Floral scents of plants are often complex mixtures of volatiles derived from several metabolic pathways. The number of presently known volatile compounds from flowers is approaching a thousand, and new compounds are constantly being discovered in various plant species. Closely related species have distinct volatile profiles that can be discriminated by specialized insect pollinators, thus limiting gene flow across species boundaries. The ability of related plant species to synthesize different volatiles in their flowers is sometimes due to differential gene expression. However, new enzymes that catalyze the formation of new floral scent components are constantly evolving through the process of gene duplication and divergence, allowing for the majority of floral scent divergence. It has been shown that very small changes in protein sequences can lead to the ability of the changed enzymes to catalyze the transformation of drastically different substrates, leading to the production of new floral scent compounds. Such evolution is facilitated by the presence of large gene families in plant genomes that already encode similar enzymes with slightly different substrate specificities; the large number of such genes makes genes duplications even more likely to occur.

Herbivore-induced plant volatiles identified from interactions at higher trophic levels: persistent effects on plants in the laboratory & field.

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In many cases, volatiles produced by plant defensive mechanisms during herbivory act directly to repel insect attack, and may have indirect effects by attracting predators and parasitoids of the pest insect. *cis*-Jasmone occurs as a component of flower volatiles, but can also be produced by damaged vegetative tissues. In an olfactometer bioassay, *cis*-jasmone was repellent to alatae of the grain aphid, *Sitobion avenae* (Homoptera: Aphididae). *cis*-Jasmone is a catabolite of the stress-produced jasmonic acid, which can induce plant defence, but had previously been considered only as a biological sink for the jasmonate pathway. However, bean plants, *Vicia faba*, exposed to *cis*-jasmone vapour were more attractive to the parasitoid *Aphidius ervi*. Moreover, wheat, *Triticum aestivum*, seedlings sprayed with formulated *cis*-jasmone 24 h previously were less susceptible to attack by *S. avenae* than control plants. In field simulator studies, significantly fewer alate *S. avenae* settled on *cis*-jasmone treated plants over a 24 h period. The intrinsic rate of population increase, r_m, of *S. avenae* apterae was reduced on *cis*-jasmone treated seedlings. In a series of small-plot experiments conducted over four years, *cis*-jasmone applications reduced cereal aphid populations infesting wheat in the field.

Polyphagy in *Helicoverpa armigera*: primary host plants, host-associated volatiles & neurosensory stimulation.

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Headspace volatiles of a subset of known host plants and non host plants of *Helicoverpa armigera* were screened by the use of Gas Chromatography linked with Electroantennography. Pigeon pea (*Cajanus cajan* L.) is one of the most attractive hosts to *H. armigera* in India, so was included as the primary reference plant to compare the presence and concentrations of electrophysiologically active chemicals across the test plants. Eight active compounds were identified in pigeon pea headspace volatiles in relatively higher concentrations. They include three green leaf volatiles ((E)-2-hexenal, Z-3-hexenylacetate and Z-3-hexenyl-2-methylbutyrate) and five monoterpenes (α-pinene, β-myrcene, Limonene (E) – β-ocimene, and linalool). The other known host plants have a small subset of these chemicals, whereas the non host plants contained even fewer. Green leaf volatiles were prominent in the host plants. The Australian *H. armigera* individuals used in this study did not respond to the group of sesquiterpenes present in pigeon pea headspace, and thus differ from their Indian counterparts. The physiological status and the sex of the moths had no effect on their responses to these chemicals. The study exemplifies how some host plants species are primary targets for searching moths, whereas the other host plants are likely to be incidental targets.

Experimental evidence of semiochemicals modulating the behavior of *Loxosceles intermedia* Mello-Leitão, 1934 (Araneae: Sicariidae).

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There is evidence of semiochemicals modulating the behavior of many species of spiders, but few reports regarding the isolation, chemical identification, and synthesis of semiochemicals in these arthropods. Our search is experimentally focused on the occurrence of volatile semiochemicals modulating the behavior *of L. intermedia*, a venomous spider that constitutes a serious public health problem in Curitiba, Brazil (3,334 people bitten in 2002). An olfactometer of rectangular arena was used. The individuals employed as stimuli were put in metallic cages, positioned against air flow. We tested all possible combinations of males and females as stimuli or test individuals. For each stimuli trial different individuals of *L. intermedia* were used. Tests were made on summer and winter 2003, and summer 2004. Only females tested during summer significantly attracted males, indicating a volatile pheromone produced during the summer season. Pheromone glands are not described for spiders, but we suppose the secretory cells are less active during winter. This is the first evidence of semiochemical in the *Loxosceles* genus. Studies involving the extraction and GC-MS identification of the putative semiochemical(s), as well as further experimental observations are being carried out.

Evolution of pheromone biosynthesis in moths.

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In addition to their role in metabolism, acyl-CoA desaturases in moths play a key role in female sex-pheromone biosynthetic pathways. These atypical membrane-bound sex pheromone desaturases have evolved a wide range of regio- and stereospecificities. Recently, it was shown that changes in pheromone blends in moths of the genes *Ostrinia* result from unusual molecular mechanisms that influence the expression and evolution of sex pheromone desaturases. In this talk, an overview of sex pheromone desaturase evolution is presented that covers the phylogeny and evolutionary patterns of sex pheromone genes, the moth sex pheromone molecular clock, and genomics of sex pheromones.

Dynamic irritability in parasitoids.

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Parasitoids may encounter other organisms while foraging on host-containing patches. Some of those organisms post direct threats to the foraging wasp. On the other hand, the patch provides resources that that can directly contribute to the parasitoid's reproductive success. Thus, the parasitoids must "decide" how to tradeoff threats versus benefits. Further complicating matters are several parameters that can affect costs and benefits, including: (1) the threatening organism may or may not attack, (2) the patch will diminish in value over time and (3) forager state, in particular, egg load, body mass and energy state. For example, heavier wasps may be less agile or slower in escape than light individuals. Here I consider 2 mutually exclusive decisions, continue foraging vs. abandon the patch as a function of the three aforementioned parameters. In my talk I will develop state-dependent theory and test some predictions from the theory as to conditions under which parasitoids will abandon patches following encounters with threats. I will show both theoretically and experimentally that sensitivity to threats are dynamic i.e. they depend upon parasitoid internal state and perception of danger.

Herbivore-induced volatiles: variability in production & variability in natural enemy response.

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Ecological interactions between plants and the enemies of their enemies are often mediated by information available as blends of volatile chemical compounds. Because the information is under control of the sender and the receiver cannot check its validity from a distance, the receiver has to base its decisions on previous experiences and this creates opportunity for the sender to cheat. Hence, we expect conflicts of interest between the sender and the receiver. We will present recent evidence for plants that cheat, i.e. produce amounts of chemical information that does not reflect the number of herbivores, so that the predators fail to select plants harbouring the highest densities of their prey. We will also present evidence showing that predatory arthropods can only partly cope with this plant-generated variability by associative learning. Based on theoretical models of the evolution of chemical languages and classic predator-herbivore-plant models extended with an information-perception interphase, we predict that the emergent non-linearities in plant-predator communication have dramatic consequences for the evolutionary and ecological dynamics of tritrophic interactions.

Influence of temperature on mechanosensory host location by vibrational sounding in parasitic wasps.

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Although temperature is one of the most important environmental parameters, there are relatively few studies dealing with its influence on ecological processes between trophic levels, e.g. on host-location mechanisms of parasitoids. Certain parasitic wasps use self-produced vibrations transmitted on plant parts to locate their immobile hosts (lepidopteran pupae) by resonance signals. This mechanosensory mechanism, vibrational sounding, depends both on physical cues of the environment and physical activity of the parasitoid and, therefore, should depend on ambient temperature and the likely adaptation of the insects to their environmental temperature range. We analysed such influences by choice experiments with plant-stem models in the temperate ichneumonid *Pimpla turionellae* and the tropical relative *Xanthopimpla stemmator*. Our results show a substantial effect of temperature on host-location activity and on the success of the process. Outside optimum ranges, the performance of the wasps decreases both at low and high temperatures. The tropical and the temperate species show substantial differences in their temperature range for successful host location and also with respect to their performance at equal temperatures. Using laser Doppler vibrometry in *Pimpla turionellae* we can furthermore show a modification of the self-produced vibrational signals by the ambient temperature with respect to both frequency and intensity.

Hostile behavior to neighbors induced by encounter experience in the Japanese queenless ant, *Pristomyrmex pungens.*

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Ant workers cooperate among nestmates within colonies while they perform strong hostile behavior against non-nestmates. It is likely that neighbors are competitors for adjacent resources while strangers are transients from distant colonies. It might be adaptive for a species having the powerful competitors to discriminate neighbors from strangers and selectively attack the former. I hypothesized therefore that the workers can distinguish members of neighboring colonies from those of distant colonies based on some presumable cues in the ant *Pristomyrmex pungens.* To test this hypothesis, I created neighbors and strangers by an experimental method and compared the ant's hostile behavior toward the neighbors and strangers. The ants which had direct encounter experience with the neighbors could distinguish the neighbors from the strangers and showed more strong hostile behavior toward the former. The recognition of the neighbors was maintained for at least ten days after the most recent encounter. Nestmates without the encounter experience showed no hostile behavior toward the neighbors. This finding suggests that information about neighbors (probably colony odor) is not transmitted to nestmates within colonies. Aggressiveness toward neighbors therefore appears to be caused and maintained on the basis of individual memories from direct contact.

Infrared sensory systems in pyrophilous buprestid beetles.

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Jewel beetles of the genera *Melanophila* (distributed nearly all over the world except Australia) and *Merimna* (with the only species *atrata*, endemic to Australia) approach forest fires. The reason for this pyrophilous behaviour is that their xylophagous larvae only can develop in burnt wood. Both genera have infrared (IR) receptors to cope with their thermal environment. However, IR receptors are totally different in *Melanophila* beetles and in *Merimna atrata*. In *Merimna* 1 to 3 pairs of IR receptor can be found on the ventrolateral parts of the abdominal sternites. Receptors are innervated by a large multiterminal thermoreceptive neuron and function according to the bolometric principle. In *Melanophila* beetles, two pairs of metathoracic pit organs can be found directly behind the coxae of the middle legs. Each pit organs houses about 70 cuticular sensilla which function according to a photomechanic principle. Physiological and behavioural data suggest that *Melanophila* beetles may use their IR receptor for the detection of remote fires. On the other hand, *Merimna* may use its receptors in the near range to prevent to land on a hot surface. The fundamentally different structure and function strongly suggest that IR receptors have evolved independently in the two pyrophilous buprestid genera.

Fire recognition on the basis of insect olfactory systems.

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Insect antennae are morphologically and physiologically highly specialised organs for odour perception. Their sensitivity, selectivity, and response time cannot be matched by any technical method of trace analysis. Utilisation of these unique abilities of insects for trace analytical purposes demands a thorough adaptation of technical devices to the special needs of insect antennae. Yet approaches using electroantennogram (EAG) techniques are highly sophisticated, however, detailed examination of sensitivity and cross-reactivity of antennal responses to binary mixtures of odours are needed. By providing these data, practical applications of biosensors on the basis of intact insect antennae as a sensitive odour detecting and recognizing device become more feasible. Besides sex pheromones, insect antennae are able to detect a broad range of odours specific to their habitat. The Black jewel beetle (*Melanophila acuminata*) as well as the Australian pyrophilic beetle *Merimna atrata* shows a very high sensitivity to wood-fire specific volatiles. The results of first field tests of the biosensor will be reported. Practical application and technical development of the biosensor in fire early warning systems will be discussed.

Fire recognition based on new technologies.

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In Europe, huneds of human beings are killed and huneds of millions € damage to property are inflicted by fires in industrial, domestic or forestry areas every year. In many cases, losses of lives and property could have been prevented by efficient fire recognition and early warning systems. For this reason, development of fire recognition systems is a rapidly growing field integrating techniques as IR/vis- detection, inorganic and organic trace gas detection and aerosol analysis. Driven by results of biological research about fire recognition mechanisms used by animals, technical research and development was focussed to IR detection and organic trace gas detection i.e. "sensors for burnt smell". Especially tin oxide sensor elements showed favourable characteristics. Their high sensitivity and selectivity to hydroxylated and phenolic compounds in combination to low cross-reactivity with water and carbon monoxide mimics highly efficient biological fire detection systems. For sensor evaluation the HRGC/SOMMSA (High resolution gas chromatography/selective odorant measurement by multisensor array) approach has been established.

Pheromone trapping of *Halyomorpha halys* in the eastern USA.

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The brown marmorated stink bug, *Halyomorpha halys* (Heteroptera: Pentatomidae), is an introduced species in the USA; first being collected in eastern PA in 1996, and subsequently spreading to NJ and MD. We have observed this bug feeding on fruit, vegetables, soybeans, and many ornamental plants. Studies were initiated to evaluate the effectiveness of attracting and capturing *H. halys* in pheromone baited traps. Various purities of synthetic *Plautia stali* (2E,4E,6Z-1:COOMe) and *Thyanta pallidovirens* (2E,4Z,6Z-1:COOMe) (Heteroptera: Pentatomidae) aggregation pheromones were loaded onto rubber septa (2.5 mg/septum) along with a UV inhibitor and anti-oxidant. The first test was started on 12 September 2003. Individual treated septa were placed inside clear football-shaped traps that were hung in fruit trees near known *H. halys* infestations. *Halyomorpha halys* male and female adults and nymphs were captured in traps baited with *P. stali* or *T. pallidovirens* pheromone. A second test consisting of various purities (76, 88, and 94% pure) of *P. stali* aggregation pheromone and 2 types of traps was initiated on 6 October 2003. Football traps with the 76% purity pheromone captured more *H. halys* than traps with the 88 and 94% purity pheromone.

Chemical ecology of the house dust mite: a potential alternative to their control?

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House dust mites (HDM) *Dermatophagoides farinae* and *Dermatophagoides pteronyssinus* are a significant health problem; their faeces are highly allergenic and can cause atopic diseases such as asthma, rhinitis, and dermatitis. Up to 85% of people with asthma in the UK (4.3 million) are sensitive to HDM allergens. Chemical acaricides and regular vacuuming are recommended to control HDM. However, areas which have been treated for HDM populations are frequently re-colonized. There is a basic understanding of HDM behaviour, but very little is known about their chemical ecology. Once identified, HDM-derived semiochemicals (behaviour-modifying chemicals) may be used as an alternative novel approach to control HDM populations in the home by manipulating their behaviour. Potential semiochemicals were extracted from *D. farinae* and *D. pteronyssinus*, and analysed using gas chromatography (GC), and mite-specific peaks were tentatively identified using coupled GC-mass spectrometry (GC-MS). A suitable bioassay was developed to test the mite-derived extracts. A Y-tube olfactometer was modified to accommodate the HDM, which has not previously been reported as a bioassay method. Extracts and components of these extracts were tested and behavioural activity was demonstrated.

Contextual control of learning in insects.

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Insects are very adept at learning the association of many types of stimuli with biologically important events such as the occurrence of food, a mate, or a predator. There are several behavioral mechanisms associated with this type of learning, and they all in general allow animals to prepare for the occurrence of the event – e.g., flee, fight, or feed. Insects are equally adept at learning, through two or more means, to ignore stimuli that are not associated with important events. This prevents overuse of limited processing capacity associated with attention. With either type of learning, single stimuli are rarely perfectly reliable. The probability of an event (e) given the occurrence of a stimulus (s) - $p[e|s]$ - might be above 0 but well below 1. But several stimuli together as a "context" might provide a more reliable predictor of an event. Associations among stimuli might underlie sensitive contextual control of a response. For example, in the context of S2 $p[e|S1]$ might be much closer to 1 than in the context of S3. Furthermore, the meaning of S1 might be entirely different in S2 and S3. The presentation will review evidence for this type of contextual control of learning in insects.

Optimal patch leaving decision rules for parasitoids in patchy environments: an individual based model.

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Experimental results have shown that when foraging for hosts in heterogeneous environments, parasitoids use a wide diversity of patch-leaving decision rules. More than the environmental stimuli implicated, it is the way they are integrated that appears as a major component of this diversity. Previous theoretical models suggest that simple rules based on environmental cues can be optimal, and that resource distribution strongly influences which is the optimal rule. However, these results were obtained considering very specific environmental conditions. Others authors claim that strong interactions exist between the parasitoids' ability to assess host availability when arriving on a patch and the best way to use the information subsequently collected while foraging in this patch. We have built an individual-based model in which parasitoids use realistic patch-leaving decision rules. Considering a large range of scenarios (depending on host distribution, environment size, parasitoid density, etc), we identify the kind of rule that is adaptive in each case. This allows us to test and discuss the robustness of previous theoretical results and to confirm or invalidate hypotheses inferred from experimental work. We show that environmental conditions have a determinant influence not only on the reliability of the initial information but also on the best use of other sources of information.

Carrion mimicry in the dead horse arum.

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Some plants rely on deception for pollination. These plants typically mimic a favourable insect resource. Attracted insects are subsequently exploited as unrewarded pollinators. Here we describe the multisensory targeting mimicry of the dead horse arum, *Helicodiceros muscivorus*. The arum produces a characteristic odor, most similar to a carcass. Odor collections from the plant and from carrion contain the same active odor components, eliciting identical antennal responses in blowflies. Addition of these odor components to scentless flowers restores their attractiveness to the same level as plants with a natural scent. The arum is furthermore thermogenic, i.e. it produces heat. The arum is capable of producing over 10°C above ambient. Although, heat production is a fairly well known phenomenon, few studies have provided direct experimental evidence as to what function heat plays. We have shown that the heat in the arum primarily serves as a behavioral altering stimulus, enhancing threefold the likelihood that the flies will enter the trap chamber of the plant. Accompanying the olfactory and thermal cues are also visual and tactile cues, reinforcing the overall carrion appearance. The dead horse arum serve as a striking example of how a plant during evolution can acquire the ability to mimic multiple aspects of complex objects.

Pheromone & visual factors contribute to catch of *Synathedon tipuliformis.*

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Currant clearwing *Synanthedon tipuliformis* (Sessiidae) has been a pioneering and successful target of mating disruption in New Zealand, widely adopted since c. 1990. However, control failures led us to examine pheromone loading and coloured traps. Male attraction to yellow delta traps was equivalent to green delta traps but was greater than to red, black, blue or white traps. Fully yellow delta traps were more attractive than black traps with yellow stripes designed to mimic the insect, and it is suggested that yellow provides a supernormal stimulus (female abdomens have two yellow stripes). However, there was no catch in the absence of the pheromone lure, showing that colour alone was not a sufficient stimulus. Catch increased as a function of pheromone loading and trap colour, but the effect appeared to be additive. However, when plastic funnel traps were used, catch in yellow traps was highly significantly greater than in green plastic traps, indicating greater discrimination between these colours. Curiously, a composite coloured lid-funnel-base of green-yellow-white funnel traps was more attractive than fully yellow-yellow-yellow coloured funnel traps or composite green-yellow-green. These results suggest that the insect has a high degree of visual discrimination between colour shades and surface area. While significant improvements (c. 16-fold) to catch have now been achieved, with an additive combination of trap colour and pheromone loading, more work is needed to decipher the visual discrimination in this species.

Putative identification of the contact sex pheromone of the burnt pine longhorn, *Arhopalus tristis* (Coleoptera: Cerambycidae).

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The longhorn *Arhopalus tristis* (F) is a pest of pines, particularly those damaged by fire, and is a major export quarantine issue in New Zealand. In our project to develop an attractant to help in monitoring and management of this species, we have investigated the contact sex pheromone. Male burnt pine longhorn initiates copulatory responses to females only after sensing them with their antennae, providing evidence for contact sex pheromone in this species. When applied to a gelatine capsule, hexane wash of female's elytra elicited male copulatory attempts similar to the copulatory response elicited in males by live females. GC-MS analysis of male and female elytra extracts indicated that all of the compounds present in both extracts were saturated and branched hydrocarbons from 25Hy to 33Hy. The GC-MS analysis of the hexane extracts of male and female elytra revealed that the profile of compounds in both extracts was qualitatively similar, but different in the ratio of two peaks. These two peaks were identified as branched 28Hy hydrocarbons and a straight chain 29Hy. Further work is underway to test the activity of these compounds.

Genetic variation in learning ability may mask population-level effects of experience.

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Variation in learning abilities has long been suspected to have a genetic basis and recently experimental evidence in support of this hypothesis is accumulating. In spite of these developments, in many studies of cognitive abilities such a variation is not controlled for, because populations of mixed or unknown genetic background are tested. We postulate that such an approach may confound the occurrence and/or magnitude of learning effects. We support this postulate with our results on learning in the predatory mite *Phytoseiulus persimilis*. *Phytoseiulus persimilis* responds to volatile chemicals released by plants following infestation by its prey, the two-spotted spider mite. The blend of induced volatiles is herbivore-specific and as such it serves as a signal of prey presence. It has therefore been predicted that learning of volatiles would be highly beneficial for this mite. We show that a collection of *P. persimilis* from natural populations on Sicily exhibits no significant change in responses to the volatiles following the experience. The same tests, however performed on iso-female lines from those populations show variation in learning ability among the lines. These as well as other results on variation in learning abilities raise the question what mechanism maintains such a variation.

Specificity of herbivore-induced volatiles in information transfer in multitrophic interactions.

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When infested by diamondback moth (DBM) (*Plutella xylostella*) larvae, cruciferous plants emit volatiles that attract a specialist parasitic wasp of DBM larvae, *Cotesia plutellae*. The wasps are not attracted to volatiles emitted by a crucifer plant infested by nonhosts [cabbage butterfly (CB) (*Pieris rapae*) larvae]. We identified specific volatile compounds emitted by cabbage plants with minor DBM larvae infestations, but not emitted by intact cabbage plants. When we compared the attractive properties of each chemical on an intact cabbage plant (treated plant) with that of the solvent alone (control plant), the wasps were equally distributed between the two plants. However, when a mixture of all the chemicals was used instead of each chemical alone, the wasps showed a significant preference towards the treated plants. These data suggest that a specific blend is important in the interaction between cabbage plants, which are infested by DBM larvae, and *C. plutella*. By comparing this tritrophic system with one consisting of cabbage plants, CB larvae, and their parasitoid *Cotesia glomerata*, the specificity of herbivore-induced volatiles in information transfer is investigated.
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Cone thermogenesis & volatiles associated with specific pollinators of *Macrozamia* cycads.

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Macrozamia cycads, endemic to Australia, are associated with an unusual and diverse array of insect pollinators compared with other cycads. Some *Macrozamia* species are pollinated by *Tranes* weevils only, others only by thrips (*Cycadothrips)*, and some by both insect types. We show that these cycads' cones have chemical and metabolic traits that are associated strongly with their specific pollinator type. Species pollinated by *Tranes*- only have cones that heat and release volatiles during the early evening, whereas *Cycadothrips*-only cycads have cones that heat and release volatiles during the day. Cones of *Tranes*-pollinated species release volatiles comprising a few monoterpenes, primarily linalool and much lower levels of beta-myrcene, whereas "thrips-only" cycads lack linalool and release numerous monoterpenes, dominated by beta-myrcene emitted at very high rates. Insect movement between male and female cones occurs during the cone heating and volatile emission phase of their specific host. Preliminary evidence suggests that insects are attracted to their host's cone volatiles and they respond neurologically to specific components within the volatile mix.

Cuticular hydrocarbon patterns of termitophiles & their host.

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Termitophile can be defined as any animal other than a termite that is normally found in a termite nest. Termitophiles supposed to use "appeasement chemicals" to integrate into the social structure of termite colonies. Since cuticular hydrocarbons are caste- and species-specific, these mixtures may important to termite, possibly used as species recognition cues. Termitophiles were collected from termite nest or around termite galleries in Taiwan. Using gas chromatographic analysis to compare the cuticular hydrocarbons of some termitophiles and their hosts, the chromatograms show some cuticular hydrocarbon components of termites might be similar to those of their termitophiles. Termitophiles might utilize chemical mimicry to integrate into termite society. The similarity of cuticular hydrocarbon component between termitophiles and termite host might indicate the integration degree of termitophiles.

Perspectives for time allocation studies in the future.

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The allocation of foraging time in insects has been a fruitful area blending empirical and theoretical research. About 100 papers with insects or mites as study subjects have appeared since 1990, the same number as for all other organisms (birds, fishes, mammals and gastropods). A survey of this literature and recent developments point to the following promising areas of research.
(A) Recent models on group foraging have not yet seen empirical tests
(B) Individual variation in behaviour has been amply documented. Three explanations have been forwarded to explain this variation: (1) information acquisition and learning, (2) differences in internal state and (3) genetic variation within and between populations. However, the relative importance of these sources of variation has not been investigated.
(B) Individual variation in behaviour has been amply documented. Three explanations have been forwarded to explain this variation: (1) information acquisition and learning, (2) differences in internal state and (3) genetic variation within and between populations. However, the relative importance of these sources of variation has not been investigated.
(C) The recent finding that symbionts, like bacteria and viruses, can change parasitoid behaviour raises the question, of which behaviours might be extended phenotypes of these symbionts, and to what extent conflicts between symbionts and their hosts occur.
(D) Host foraging, the most studied process in patch allocation is one facet of different trade-offs, such as searching for food and searching for hosts. Little is know on how these trade-off affect parasitoid foraging time and how the animals integrate the information of the presence of different resources.
(E) Up to now the study field of patch residence time is restricted to behavioural macroscopic, processes. Insect physiologist should be attracted to the field to unravel processes as the integration of different signals, how the time elapsed is integrated to patch leaving mechanisms, etc.

A maternal chemical cue affecting the behaviour of a parasitoid of aphids on emergence.

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A fully-defined artificial diet containing no secondary plant compounds produces aphid mummies which give emerging parasitoids no opportunity to learn plant volatiles of the plant on which they were reared. To improve parasitisation of the aphid *Myzus persicae* (Sulzer) by the generalist parasitoid *Aphidius colemani* (Viereck) on the diet, we allowed parasitisation on a host plant leaf for 48 h, after which the aphids were returned to the diet. We now needed to test that this procedure did not provide plant volatile cues to the emerging parasitoids. Unfortunately it did, but only when the plant from which the parent parasitoids had been reared was the same as that on which they had parasitised the aphids. Thus, if parasitisation has occurred on Brussels sprouts, the emerging daughters only preferred the odour of sprouts to cabbage if their parents had been reared on sprouts; if reared on cabbage, there was no olfactory preference for either brassica. The parasitoids clearly leave a maternal cue for their offspring when they oviposit, and we discuss what this might be, where it might be located in the parasitised aphid, how it affects the emerging offspring and why it is left.

Sexual behavior in *Nezara viridula* (L) (Heteroptera: Pentatomidae).

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The southern green stink bug *Nezara viridula* (L.) (Heteroptera: Pentatomidae) has a complex sexual behavior. As a part of a specific mate recognition system, during courtship male and female exchange chemical and substrate-borne vibrational signals. Sexually mature males release a volatile sex pheromone that is regarded as a long-range attractant and serves to bring bugs to the same plant. When on a plant, both male and female emit several species and sex specific vibrational signals called songs. It seems that volatile chemical signal triggers the emission of the female calling song. Male responds to female calls with his songs and he locates the singing female on a plant using her vibrational signals as a directional cue. Male songs keep female singing for a longer time with a regular repetition rate while the female calling song modulates the emission of the male sex pheromone. Such feed-back modulatory mechanisms probably help to improve the synchronization of sexual activity and could also provide the sensory cues for sexual selection.

Mating behaviour of *Zorion guttigerum* (Westwood) (Coleoptera: Cerambycidae).

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Zorion guttigerum *(Westwood) is a diurnal cerambycine beetle endemic to New Zealand. Pair-bonding, mating, and adult feeding occur on flowers while larvae feed on dead tree twigs. Adults live for > 3wks and the male and female arrive at flowers independently, suggesting that long-range sex pheromones may not operate in mate location. Mean pair-bonding period is 22.7 ± 24.2 (mean ± SD)(ranged from 2.3 to 120.5) min during which time 1.8 ± 1.5 (ranged from 1 to 6) matings occur. Mating lasts an average of 4.7 ± 2.5 (ranged from 1.2 to 11.7) min. Overall operational sex ratio is male-biased (»1♀:1.5♂) but it becomes very highly male-biased (»1♀:9♂) when pair-bonding and feeding activities decrease to the minimum between 1400 and 1600 h, suggesting that females leave flowers for oviposition. It is probably true that for cerambycid species whose females oviposit alone and mating and oviposition occur on different plants or different parts of plants, the operational sex ratio varies significantly over time on the mating/feeding sites. Duration of pair-bondings also varies over time. Shorter pair-bondings in the morning may suggest a strong sexual selection process. After ca. 2-h selection, both sexes tend to engage in much longer pair-bonding and mate more times before females leave the mating and feeding sites for oviposition in the early afternoon.*

Towards the identification of contact pheromones of the fig longicorn, Agrianome spinicollis *(Macleay).*

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Agrianome spinicollis is a cerambycid endemic to the eastern states of Australia and is a serious pest of pecan nut trees. Damage results from larvae feeding within the limbs and trunks of the trees, which ultimately results in the trees' collapse. Observations suggest that contact pheromones on the elytra of female beetles form an important component of the mate recognition system employed by male beetles. Experiments were conducted in which the cuticular hydrocarbons were removed from both male and female beetles. Such solvent-rinsing of females resulted in the loss of attractiveness towards males. The subsequent re-application of the hexane rinse, at concentrations as low as 0.1 female equivalents, was followed by mating attempts by males. Gas chromatographic analysis of the cuticular rinses revealed significant quantitative sex differences in shared components, and possibly qualitative differences. The male extract is dominated by mono-unsaturated hydrocarbons having 25, 27 and 31 carbons. The female extract, in contrast, is dominated by a single alkene, with 26 carbon atoms. As a result of the short range of efficacy of the compounds, these findings are unlikely to have any significant management implications.

Influence of lek size on female visitation in Queensland fruit fly (*Bactrocera tryoni* Froggatt) (Diptera: Tephritidae).

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The lek mating system of Queensland fruit fly, *Bactrocera tryoni* (Froggatt) (Diptera: Tephritidae), may reduce the ability of this species to colonise new areas. The importance of lek size for female visitation at lek sites and initiation of male pheromone-calling was investigated in large cages using artificial leks. Female visitation at artificial leks was monitored to determine if females visited leks due to passive attraction or showed a preference for larger leks. Female *B. tryoni* display a preference for larger leks rather than being passively attracted to a greater perceived stimulus from large leks. Per capita success of males in attracting females was higher at larger leks than where there was a single male. Calling propensity of male *B. tryoni* was increased by the presence of conspecific males. Overall, female visitation at leks was only weakly associated with male calling, suggesting that lek size and the number of pheromone-calling males is not the only factor important in locating mates in this species.

Integration of multi-sensory information in orientation & navigation behavior.

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The ultimate goal of neuroethology is to describe the neural mechanisms that produce adaptive behaviors in freely moving animals. My lab has been using a virtual reality flight system that closely emulates realistic stimuli that occur during flight, and which produce robust flight behaviors in a tethered moth (*Manduca sexta*) that are similar to those seen in real flight. Concurrently we are able to utilize extracellular multi-unit electrodes to record activity in the region of the ventral nerve cord between the brain and the thoracic ganglia. This system allows us to investigate mechanisms by which *Manduca* responds to visual and olfactory stimuli that it may encounter during actual pheromone-guided flight, and how it might activate and control flight manoeuvres. Initial results reveal ensembles of neurons that show unique activation patterns associated with visual pattern movement associated with flight, and that these patterns are modulated during exposure to pheromone. Similarly, other unique ensembles of neurons are activated differentially when the preparation is in tethered flight or not flying. These experiments are aimed at understanding how multiple streams of information are integrated by the nervous system and how this is meshed with ongoing behavior. Supported by: ONR-MURI No. N00014-01-1-0676.

Pheromone-modulated orientation mechanisms in walking male cockroaches.

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Perhaps the most remarkable olfactory task achieved by insects is that of locating distant resources using airborne chemical cues. It is currently thought that two primary mechanisms underlie flying insects' ability to perform this behavior: 1) an odor-modulated, visually-mediated orientation to the wind direction and, 2) an odor-activated internal program of turns. The blending of these two mechanisms is thought to generate the upwind zigzagging paths that we observe from insects tracking odors in flight. It is not yet clear whether vision contributes significantly to the odor tracking behavior of walking insects. Recent results suggest that visual information may be very important, but not used in the same manner as flying odor tracking insects. Furthermore, the structure of the walking paths of pheromone tracking cockroaches look remarkably like those observed from flying insects, and thus may be governed by similar mechanisms. Our new results suggest that comparing the odor tracking behavior of walking and flying insects in similar environments may reveal important similarities and differences in how olfactory and other sensory information is used. Supported by: ONR-MURI No. N00014-01-1-0676.

Odor concentration & odor invariance in the olfactory system of the honeybee.

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Sensory systems have evolved to recognize stimuli over a range of stimulus intensities. A stimulus may be more intense, but it will still retain other qualities that make it classifiable along another dimension. When the qualitative representation of a stimulus remains the same over a specific range of intensity, the stimulus is defined as *invariant* over that range. In olfaction, it is often noted that increasing the intensity of an odorant affects its quality, such that odorants do not remain invariant as a function of their concentration in air. I will discuss how our experiments with honeybees reflect the extent that odorants remain invariant and the implications of failure of invariance for odor coding.

Local resource competition in the solitary parasitoid *Haplogonatopus atratus* **(Hymenoptera: Dryinidae).**

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This is the first study showing local resource competition (LRC) in parasitoids. Females of the parasitoid *Haplogonatopus atratus* are apterous, while males are alate. Individuals of the host, *Laodelphax striatellus* (Hemiptera Delphacidae), are distributed in clumps in paddy fields. These facts suggest the existence of LRC among female offspring of mother parasitoids, and that female parasitoids will produce a lower proportion of male offspring in patches with higher host density. Our laboratory experiments confirmed these predictions, as follows. A female parasitoid was placed in a glass pot with rice seedlings on which different numbers of planthoppers (10, 20, 40, or 80) were feeding. The female was allowed to oviposit freely until she left the seedlings. Sex ratios (male proportions) in emerging adults decreased with increasing host density. Moreover, the average number of eggs laid among parasitized hosts decreased as the host density increased, but hosts with different numbers of eggs laid in the same patch produced similar sex ratios. Host-quality models suggesting that male eggs are more likely to be laid in hosts of low quality (i.e., the parasitized hosts in this case) do not explain the above patterns in the sex ratio.

Identification of semiochemicals for the Asian longhorned beetle, *Anoplophora glabripennis*.

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A series of long-chain hydrocarbons comprise the cuticular waxes of both sexes of ALB adults. Although for the most part the gas chromatographic profiles are similar for the two sexes, five monounsaturated compounds were consistently more abundant in samples from females than in those from males. These compounds were identified as (*Z*)-9-tricosene, (*Z*)-9-pentacosene, (*Z*)-7-pentacosene, (*Z*)-9-heptacosene, and (*Z*)-7-heptacosene in the approximate ratio of 1:2:2:8:1, respectively. Antennal contact to a micro-centrifuge tube coated with synthetic mixture of the five compounds stimulated copulatory behavior in males. This is the first time that monounsaturated hydrocarbons, (Z)-9-tricosene, (Z)-9-pentacosene, (Z)-7-pentacosene, (Z)-9-heptacosene, and (Z)-7-heptacosene, have been implicated as contact sex recognition pheromone components in the family Cerambycidae. Two male-specific beetle volatiles were found that elicited strong gas chromatographic-electroantennographic detection (GC-EAD) responses from both sexes of adult Asian longhorned beetle (ALB), *Anoplophora glabripennis*. The secretion consisted of a ~1:1 blend of functionalized dialkyl ethers, 4-(*n*-heptyloxy)butanal and 4-(*n*-heptyloxy)butan-1-ol. These compounds are chemically unusual natural products that are previously unknown from insects. Laboratory olfactometer studies showed that a blend of 10 µg of each synthetic compound on a filter paper strip was significantly attractive to ALB adults.

Identification of sex pheromones of ***Holcocerus hippophaecolus*** **(Lepidoptera: Cossidae) & biological activities.**

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Holcocerus hippophaecolus Hua (Lepidoptera: Cossidae) is a destructive forest pest widely distributed throughout northern and western region of China. Its primary host-plant is sandthorn (*Hippophae rhamnoides* L.), which is a most widely planted shrub used for preventing from soil erosion and desert control, along the area of loess plateau in China. The fruit of sandthorn could be used for juice and Chinese medicine. Its plantation reached 180 million ha. About half million ha of plantations suffered severely attacking by the pest insect. Seven components were identified from female sex pheromone gland. These components were (*E*)-3-tetradecenyl acetate (E3-14:Ac), (*Z*)-3-tetradecenyl acetate (Z3-14:Ac), (*Z*)-7-tetradecenyl acetate (Z7-14:Ac) and related alcohols: E3-14:OH, Z3-14:OH, Z7-14:OH and (*Z*)-9-tetradecenyl acetate (Z9-14:Ac). Two major ones are as Z7-14:Ac and E3-14:Ac. EAG showed that Z7-14:Ac produced the strongest response and followed by E3-14:Ac in these seven compounds. The field trials showed that the combination of Z7-14:Ac and E3-14:Ac in a ratio of 1:1 could significantly catch much more males than others.

Different habitats, different habits? Information use in the parasitic wasp *Venturia canescens*

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Different environments require different adaptations. Effective foraging strategies in one habitat can lead to a wastage of time or energy in another habitat. In the parasitoid *Venturia canescens* (Gravenhorst), wasps with two different reproductive modes occur under different ecological circumstances. We studied the use of foraging information by testing females from ten populations in multi-patch experiments. Only thelytokous (unisexual) females, that usually live in habitats with fluctuating host densities, readily responded to cues indicating the current abundance of host patches by a reduction in patch residence times. In contrast, no changes in patch time allocation could be detected when we tested arrhenotokous (bisexual) wasps from environments that are more stable with respect to host density. Additional experiments showed that the different response patterns were not produced by different learning abilities of the two wasp types but instead by different ways of how the information gained was dealt with by the searching females. From supplementary life history data we conclude that the observed differences in information use in *Venturia canescens* are more likely to have evolved in response to life history-specific needs of dispersal in arrhenotokous wasps that are irrelevant to thelytokous wasps rather than to habitat-specific fluctuations in host abundance.

Maternal care of 8 Costa Rican Weevils (Attelabinae, Attelabidae, Coleoptera)

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An ethological study was carried out in Costa Rica during the period from April 1999 to March 2000 in order to reveal the maternal behaviours of attelabin weevils endemic to Central and South America. The whole process of the cradle formation was analysed for 8 species of 4 genuses (*Euscelus elliptiguttatus* Voss, *E.* sp.1, *E.* sp.2, *Omolabus quadratus* (Sharp), *O. angulipennis* (Sharp), *Xestolabus conicollis* (Sharp), *X. corvinus* (Gyllenhal) and *Pilolabus rugiceps* Voss). *E.* sp.1 and *E.* sp.2 made two types of cradles; cradles of one type made of the periphery of a leaf showed similarity with the cradles of *Euops*. *X. corvinus* firstly cut at the basal part of a leaf as many species do, and secondary at the apical part it conducted additional cutting with which the apical portion were thrown away. Leaf rolling technique of *P. rugiceps*, which made spherical cradles, was unique and it appeared to be a set of restricted manner. I am grateful to INBio and the staff (especially A. Solis, C.M. Rodriguez and Q.M. Jimenez) for their help and for giving me an opportunity to study at INBio. I also thank Dr. D.H. Janzen for valuable information and Dr. W. Eberhard, Dr. P. Hanson and K. Nishida for their kindness.

The Pherobase: database of insect pheromones and semiochemicals

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The Pherobase is a database of insect pheromones and semiochemicals that has become available on the worldwide web since November 2003. The main objective of this database is to provide a guide to the literature published on chemicals signals involved in insect's chemical communication. The pherobase is not biased towards any insect order or any type of semiochemicals; it covers most insect orders and all types of semiochemicals. The database is extensively linked, user-friendly, easy to understand and to follow. Currently, there are over 5000 entries that cover most of the insect orders, around 1600 molecules, and over 16000 static html pages. In addition, mass spectral data collected from the literatures for more than 800 compounds are included. The Pherobase will be demonstrated electronically in this meeting and I would appreciate any inputs form Entomologists on its features, structure and how to proceed with it in the future?

Role of leaf volatiles from two maize (Zea mays L.) cultivars in eliciting larval orientation of Chilo partellus (Swinhoe) (Lepidoptera: Pyralidae)

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The effects of two maize cultivars, susceptible (Basilocal) and resistant (Kisan), and their methanol- and hexane- extracts in eliciting orientation of the first-instar Chilo partellus (Swinhoe) larvae were studied in the laboratory. When presented at one end of the orientation tunnel, leaf whorls of both the Basilocal and Kisan varieties, elicited higher orientational responses of stemborer larvae as compared to the blank end, the orientation preference (OP) of the larvae being + 72% and + 48%, respectively. Statistically, OP of larvae for Basilocal leaves was significantly higher than for Kisan leaves ($P<0.01$). Methanol extracts of both the Basilocal and Kisan leaves also attracted the neonate larvae, but the difference between the OP for the two was not significant. However, the attraction of larvae for hexane extractable chemicals of Basilocal (OP= 40%) was significantly higher than that for Kisan (OP = -32%). The observations indicate that difference in the orientational response of stemborer larvae for the two maize cultivars is due to non-polar semiochemicals, extractable in hexane. Identification of the chemicals in these maize cultivars can be useful in the behavioural management of this economically important insect pest of maize and sorghum.

Assessing the pitfalls of insect repellent evaluation methodologies

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Insect repellents provide useful protection from malaria, for high-risk groups including forest-workers; especially in the Amazon Basin where vectors are difficult to control through conventional means due to their exophily, crepuscular biting and forest-breeding. The WHO recommends that repellents are field-tested using human-landing catches, with individual collectors positioned >10m apart. However, it is still commonplace to perform tests using repellent treated "bait" and an unprotected "collector". This could overestimate repellent protection and longevity since mosquitoes will feed preferentially from the unprotected "collector", thus potentially exposing subsequent users to disease. Mosquito diversion from 15% deet (repellent) to 15% baby oil (control) was quantified utilising pairs of testers with each team situated >10m apart. Permutations of repellent repellent; control control; and repellent control were replicated over 18 nights with 6 pairs. Control individuals received 22.82% more bites when paired with repellent wearers, than untreated individuals (likelihood-ratio test Prob > chi2= 0.0485), and repellent wearers received 11% more bites when paired with other repellent wearers than controls (N.S.). In a further test with 25% lemongrass *(Cymbopogon citratus)* repellent, controls received 23.53% more bites when paired with repellent wearers, than with unprotected partners (N.S.). Thus it is recommended that repellents are field-tested using individuals and not pairs of collectors.

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Do sensory detectors interact in the decoding of environmental signals?

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In a multimodal context, there is presumably little or no interaction between sensory detectors. Rather, a multitude of detector phenotypes and sub-phenotypes exist to deconstruct a signal rich environment and to present these signals to the brain via diverse pathways. The brain integrates this horde of information, making use of what is interesting and discarding what is not. Or, perhaps, there is a great deal of interaction, and we've been ignoring it. Individual neurons may contain multiple receptor/transduction pathways responsive to different signals, integrating these signals at the biochemical level. Individual sensilla (sensory organs) may contain neurons which respond to different signals and when activated, alter properties of their sibling neurons. Sensory appendages (eyes, antennae, wings, legs) may contain sensory organs with modalities as diverse as the functions of the appendage itself. Developmental decisions establish these diverse detector arrays, and hormonal regulation may differentially modulate their individual activities. My presentation will focus on the potential interactions between diverse sensory detectors, striving to consider sensory detectors in ways not currently in vogue.

Male-produced anti-sex pheromone in a plant bug.

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In at least some plant bugs (Miridae) females produce sex pheromones in the metathoracic scent gland (MSG), which in most other true bugs (Heteroptera) is responsible for chemical defense. Females of a pine inhabiting mirid, *Phytocoris difficilis* Knight, contained predominantly hexyl, (*E*)-2-hexenyl and (*E*)-2-octenyl acetates in their MSG, while the latter two compounds were found only in trace amounts in males. This ternary acetate blend (2: 1.5 :1 by volume) constitutes the sex pheromone of *P. difficilis.* However, the possibility that the MSG secretion of male plant bugs plays a role other than defense has been largely overlooked. For *P. difficilis* we showed that hexyl butyrate and (*E*)-2-hexenyl butyrate are abundantly produced only in males, and demonstrated that these MSG compounds elicit strong antennal responses in conspecific males. Furthermore, physiologically relevant doses of these butyrates totally interrupted attraction of males to the female-produced sex pheromone in the field. Our results suggest that in some plant bugs the male MSG esters have a natural communicative function as anti-sex pheromones, probably to repel other males from further mating attempts. From a practical standpoint, synthetic anti-sex pheromones may be useful for mating disruption of economically important plant bugs.

Sex pheromones in Diprionidae & other Symphyta (Hymenoptera).

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The most ancient group of Hymenoptera, Symphyta or sawflies, consists of around 8000 described species. The vast majority, are herbivorous and some species are important pests of agricultural crops or forest trees. Despite this, comparatively little is known about the chemical ecology within this group. I will present an overview of the sex pheromones identified so far in the different families, with emphasis on Diprionidae, the only well-studied group. In this family all active compounds discovered are saturated methyl-branched hydrocarbons with two to four chiral centres. The chiral configuration seems to be important for all species, whereas specificity regarding carbon chain-length or functional group (different esters) often is less distinct. The electrophysiologically and behaviourally active esters are released by the females together with their alcohol and carboxylic acid precursors. According to present knowledge, several sympatric species share the same sex pheromone, also with respect to stereoisomer, calling for more attention from chemical and evolutionary ecologists. In the other families the identified pheromones are different from those in Diprionidae, but there are too few sex pheromones identified for drawing any general conclusions.

Identification of volatile compounds from *Stenodema* (Heteroptera: Miridae) trigged by their natural predators.

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Gland secretions in Heteroptera contain different compounds spanning from sex pheromones to allomones. Little is known about the structure and function of released defence substances, especially in Miridae, and studies of emissions from living species are rare. In this study, *Stenodema* Laporte specimens were exposed to predators and the trigged chemical response was recorded in vivo using solid-phase microextraction (SPME). Sexual immature adults from *S. calcarata* Fallén, *S. holsata* Fabricius and *S. laevigata* Linnaeus were tested and predators like spiders and Nabidae were introduced to provoke defence reactions. Each species of *Stenodema* released three substances in high amounts due to the confrontation. These compounds were absorbed on SPME-fibers and identified using GC-MS.

Functional diversification of the material properties of orb-weaving spider silk.

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Spider silks are exceptionally strong and can absorb more energy before breaking than high performance manmade fibers such as Nylon or Kevlar. The exceptional mechanical properties of spider silks are thought to have resulted in part from selection on the function of webs during prey capture, in particular upon aerial orb webs that must arrest fast-flying, high kinetic energy insect prey. However, orb-weaving spiders spin up to seven different types of silks, all of which are produced from distinct glands and are utilized in many different ways, from protecting eggs, to building retreats, to capture of prey. Here, we use the orb-weaving spider *Argiope argentata* to demonstrate that each of these silks, produced by distinct glands within a single spider, has a unique combination of material properties. Furthermore, these differences in mechanical performance reflect variation in the amino acid sequences of silk proteins. Thus, molecular evolution has played in important role in the evolution of the diverse toolkit of silks utilized by modern orb-weaving spiders.

Candidate pheromone receptors of the moth.

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The remarkable responsiveness of male moths to female released pheromones is based on the extremly sensitive and selective reaction of highly specialized sensory cells in the male antennae. These cells are supposed to be equipped with very distinct, male-specific receptors for pheromonal compounds; the nature of these receptors is still elusive. Using a combination of genomic sequence analysis and cDNA library screening of the tobacco budworm *Heliothis virescen***s led to the discovery of a divergent gene family encoding putative olfactory receptors. Comparing the encoded amino acid sequences of all candidate olfactory receptors emphasized their extreme diversity; however, a small family (identity > 40%) emerged, including the three receptor types HR14, HR15, HR16. RT-PCR-analysis revealed that all three subtypes were exclusively expressed in the antennae of male moths. More detailed** *in situ* **hybridization studies indicated that expression of these male-specific receptor types was confined to antennal cells located beneath sensillar hair structures which contain pheromone sensitive neurons. Moreover, two colour double** *in situ***-hybridization approaches in combination with laserscanning microscopy uncovered that each cell expressing one of these receptor type was surrounded by cells expressing pheromone binding proteins. These findings suggest that the novel receptor types may render neurons in male antennae responsive to pheromones.**

Influence of host footprints kairomone & past oviposition experience on the host location behaviour of *Trissolcus basalis.*

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During the host selection process insect parasitoid make flexible use of chemical information which can be directly or indirectly associated with the presence of their hosts. Wasp's response to a stimulus should be inversely related to its potential response and a change in wasp's response to stimuli can be the result of associative learning. In this view, influence of host indirect cues on the host location behaviour of the egg parasitoid *Trissolcus basalis* was investigated. *T. basalis* host selection behavior is mediated by chemical cues which are directly or indirectly associated with the presence of host egg mass of *Nezara viridula*. In particular, nymphs and adults footprints left on the substrate are known to play a role as indirect host-derived cues inducing an arrestment response in the parasitoid. We hypothesized that females which experienced the presence of host footprints kairomone without finding the host eggs should be less likely to respond to the same stimulus in the near future, while females that successfully located host egg masses would have shown reinforced responses. Results support our hypotheses. The significance of these findings for the manipulation of adult parasitoid behavior to enhance pest control and their implications for host selection by egg parasitoids are discussed.

Influence of direct mutual interference on patch time location in egg parasitoids.

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Direct mutual interference, i.e. the competition between parasitoid females foraging simultaneously for hosts, has regularly been considered to have a strong influence on the population dynamics of host-parasitoid systems. Furthermore, patch time allocation has also always been presented as an important behavioral component that should be deeply involved in the demographic features of host-parasitoid interactions. Based on a detailed analysis of within-patch interactions between *Trissolcus basalis* females, the present study aimed at understanding accurately how direct mutual interference can influence patch time allocation in this species. *T. basalis* attacks patches of its host, the egg masses of the Southern Green Stink bug *Nezara viridula*. In this species, competition between females occurs through strong fights when they are foraging simultaneously on the same patch. Such mutual interference could modify drastically the time invested by the females on the host patches. Behavioral interactions between the females were analyzed using a Cox's proportional hazards model. Results first indicated that the competitive interaction (through contacts and fights) between females strongly influences their patch residence time. Also, all the corresponding patch-leaving decision rules adopted by the females appeared to be under a significant genetic variation in the population studied. Such a genetic variability likely corresponds to an intra-population genetic variation in females' susceptibility to interference. The population dynamics and evolutionary consequences of these results are discussed.

Investigation of pheromones of coffee stemborers (Coleoptera: Cerambycidae).

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Stemborers (Coleoptera: Cerambycidae) are the most important insect pests of arabica coffee in Africa and Asia. In India and other countries of S.E. Asia, the main species is *Xylotrechus quaipes*. Adult males were previously shown to attract female beetles to traps in the field. Analysis of volatiles collected from virgin male beetles by gas chromatography (GC) linked to electroantennography (GC) showed three components accounting for over 90% of the volatiles, two of which elicited EAG responses from a female beetle. The major EAG-active component was identified as (*S*)-2-hydroxy-3-decanone (I) and the minor as (*S,S*)-2,3-dihydroxyoctane (II). The third component was 3-hydroxy-2-decanone (III). (*S*)-(I) was attractive to male *X. quaipes* in laboratory bioassays, but addition of (*S,RS*)-(III) at 10% of (I) reduced attractiveness. In field trials carried out in India with sticky, cross-vane traps, (*S*) and (*RS*)-(I) attracted male *X. quaipes* and addition of (*S,S*)-(III) at 10% of (I) reduced attractiveness. In Africa the main species is *Monochamus leuconotus*. Preliminary field studies have failed to show any attraction between the sexes, but collection and analysis of volatiles from male or female beetles has shown the males produce a compound not produced by the females and identification of this is in progress.

Chemical information in soil: tritrophic interactions between plants, insects & parasitic nematodes.

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Plants can protect themselves against insect herbivores by promoting the effectiveness of the herbivore's enemies. One way to do this is by releasing herbivore-induced volatiles that attract the herbivore's enemies. Most evidence for this phenomenon comes from studies on aboveground tritrophic interactions. We will discuss an example from underground tritrophic interactions: roots from a coniferous plant (*Thuja occidentalis*) release chemicals upon attack by weevil larvae (*Otiorhynchus sulcatus*). These chemicals attract parasitic nematodes (*Heterorhabditis megidis*) which are the natural enemies of the weevil larvae. We will discuss biological, chemical and exploitation aspects of this tritrophic interaction.

Collective foraging in an outbreaking forest pest.

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Gregarious behaviour is common among outbreaking forest insects. The forest tent caterpillar (*Malacosoma disstria*) is an early spring feeder that attacks North American hardwoods. Colonies exhibit complex social behaviour, using pheromone trails to move together between temporary bivouacs and feeding sites. Colonies alternate together between periods of quiescence and foraging, and mass movement is facilitated by chemical communication: moving caterpillars spin a fine thread of silk from their mouthparts and mark it with a highly stable non-volatile sterol pheromone. We show that the presence of trails significantly influences the behaviour of individual caterpillars, especially in the early larval stadia: trails increase both amount of locomotor activity and speed of travel. Larvae find novel food sources faster in an arena coated with trails than in a bare environment. This translates into a significant effect of trails on individual performance: caterpillars consume more food, grow larger and develop faster in the presence of trails. Although forest tent caterpillars do not recruit colony-mates to new food finds, trails nonetheless improve foraging by increasing individual mobility. These findings suggest that group-living is integral to the biology of the forest tent caterpillar and has shaped many aspects of its behaviour and development.

Finding the needle in a smelly haystack: prey location by carnivorous arthropods.

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Many natural enemies of herbivorous arthropods use herbivore-induced plant volatiles to locate their prey. These induced volatiles are sometimes referred to as 'SOS-signals' from plants and their composition is variable. The composition of a blend of herbivore-induced plant volatiles can vary with plant and herbivore species, and with other biotic and abiotic conditions. Carnivores have to cope with this variation to be able to locate their food. We investegated responses of a predatory mite to plant volatiles induced by its prey or by non-prey herbivores. The role of individual compounds from the volatile blend in the discrimination between different blends has been addressed, as well the importance of phenotypic plasticity in predator behaviour. Our data show that the predatory mite *P. persimilis* can locate their prey by using SOS-signals from the prey's host plant not only in setups that have no odour-background, but also when an odour-background consisting of other herbivore-infested plants is present. The induction of herbivore-induced plant volatiles is mediated by the phytohormone jasmonic acid. Application of this hormone can mimic the effect of spider mite infestation to some extent. Moreover, application of the phytohormone can increase the effect of spider mite infestations at low mite densities.

Interaction between olfactory & gustatory inputs in insect behavior.

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Behavior of insects is governed by chemical and physical signals impinging on specialized sensory organs. Volatile chemical signals are detected by the olfactory sense and result in upwind orientation by conspecifics receiving them. Detection of less volatile chemical signals by the gustatory sense results in behaviors such as copulation, biting and ingestion. However, seldom are these signals encountered in isolation; more often multiple chemical signals are detected simultaneously and the insect receiving them must process the information and appropriate a response. We investigated behavior of Colorado potato beetles challenged by simultaneous olfactory and gustatory stimuli. An open ambulatory Y-track olfactometer was used to measure insect preferences, number of turns made during orientation, and time to make a choice between stimuli. Olfactory stimuli were delivered to one-side of the device; gustatory stimuli were painted on the other side. Behavioral conflicts and preferences were discovered between stimulus pairs. For example, females took more time and made more turns when confronted with a volatile plant attractant and sucrose compared to the plant attractant and a male extract. We will examine behavioral hierarchies revealed by our studies and discuss how they may relate to reproduction and feeding.

Does the aggregation pheromone of *Riptortus clavatus* (Heteroptera: Alydidae) attract the *Piezodorus hybneri* (Heteroptera: Pentatomidae)?

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The male of *Riptortus clavatus* releases an aggregation pheromone which attracts conspecific adults and nymphs. We found that the synthetic aggregation pheromone (a mixture of three components) of *R. clavatus* (Heteroptera: Alydidae) attracted nymphs and both sexes of adults of *Piezodorus hybneri* (Heteroptera: Pentatomidae) in soybean fields. Among the three components of the synthetic pheromone, (*E*)-2-hexenyl (*E*)-2-hexenoate (EE) showed the attractiveness to *P. hybneri* and the activity was almost equal to the aggregation pheromone. The other two components lacked the attractiveness. Thus, EE was considered to be the component which showed the attractiveness to *P. hybneri*. Since we could not detect a peak of EE with the GC-MS analysis in the hexane extracts of *P. hybneri* and soybean plants, *P. hybneri* seems to utilize the component of the pheromone of *R. clavatus*, which has a similar host range, as a kairomone to search for food plants. However, real male bugs of *R. clavatus* did not attract the *P. hybneri*. Now we are trying to explain this contradiction.

Fatty acid levels & triacylglycerol biosynthesis in the sex pheromone gland of *Heliothis virescens* Fabricius.

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Most species of moths biosynthesize sex pheromone de novo in a specialized gland located between the 8th and 9th abdominal segments. As well as sex pheromone components, the gland contains large quantities of lipids, predominantly triacylglycerols. These triacylglycerols contain significant quantities of fatty acid precursors and other intermediates of the sex pheromone components. Despite the obvious role as a source of precursor acids for pheromone biosynthesis, the evidence in favor of this is equivocal; one particular problem is that insects using specific geometric isomer ratios for pheromone components tend to have very different ratios of the respective precursor acids. We believe that the glandular lipids have the role in pheromone biosynthesis of governing glandular fatty acid concentrations. When fatty concentrations are high (especially during high rate of pheromone biosynthesis), excess fatty acids are stored in the lipids; when concentrations low, fatty acids are mobilized from the lipids. This ensures the smooth production of pheromone during the sexually active period, which otherwise might be punctuated, for example, by high fatty acid concentrations inhibiting pheromone biosynthetic function. In this talk, we report the effect of glandular fatty acid levels on the biosynthesis of triacylglycerols in the pheromone gland of *H. virescens*.

Male mating behavior of the white-spotted longicorn beetle, *Anoplophora malasiaca* (Thomson) (Coleoptera: Cerambycidae) regulated by assorted female components.

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Male *A. malasiaca* dash towards and mount females, and bend the abdomen to copulate after contact with antennae or tarsi. Males make a similar mating attempt with glass dummies treated with the solvent extract of female elytra, indicating the presence of a female sex pheromone perceptible by physical contact. Chemical analysis of the pheromone components from female elytra revealed that at least three different groups of chemicals are involved: (1) long-chained alkanes, (2) ketones and (3) novel polar components. All three of these groups of chemicals are indispensable to evoke male mating behavior and work synergistically. Components within each group, however, complement each other. In addition to observation of the contact sex pheromone, we note the presence of volatile attractant produced by females in the near vicinity. Visual cues of females also appear to act synergistically with the volatiles in male mate location before direct contact by male antennae.

Control of sex pheromone precursor biosynthesis in adzuki bean borer *Ostrinia scapulalis.*

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The adzuki bean borer *Ostrinia scapulalis* is one of the eight *Ostrinia* species found in Japan, and is an important pest of adzuki bean plant and hop. The sex pheromone of *O. scapulalis* is a blend of (*E*)-11- and (*Z*)-11-tetradecenyl acetates. These components are biosynthesized from common saturated fatty acids through chain shortening, desaturation, reduction and acetylation. The key enzyme in this cascade is the Δ11 desaturase that introduces a double bond into myristic acid to form E11-/Z11-tetradecenoic acid (fatty acyl pheromone analog, FAPA). It is known that one day-old females of *O. scapulalis* do not show calling behavior. We found that these females had little FAPA in their pheromone gland while sexually mature 3 day-old females accumulated a substantial amount. As decapitated females did not synthesize FAPA, it appeared that a signal from cephalic organs is important for triggering FAPA biosynthesis. We further found that FAPA titer in the females maintained under a constant light condition is lower than that in the control females, suggesting that the light-off signal enhances FAPA synthesis. Since the expression level of Δ11 desaturase gene was constant regardless of FAPA titer, it was suggested that FAPA biosynthesis in *O. scapulalis* is under post-transcriptional regulation.

Pollinator responses to mimicry by sexually deceptive orchids.

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Deceptive orchids mimic insect signals to attract pollinators without providing a reward. Orchid flowers may resemble food, oviposition sites, or female insects. Insects that respond, by attempting to mate with or feed from orchid flowers for example, collect and distribute orchid pollinia. Deceived insects may incur costs, e.g. time and energy wasted travelling to and interacting with orchids, and loss of sperm during pseudocopulation. Such costs may encourage the evolution of behavioural strategies that allow insects to avoid deceptive orchids. In sexually deceptive orchids, male pollinators are initially attracted by scents identical to female insect sex pheromones. Short-range cues eliciting pollinator contact with the flower may include visual cues such as colour and shape, and structural or tactile features that resemble female insect bodies. Investigation of the relative importance of chemical, visual, and structural cues, and the interaction between, long- and short-range cues should be rewarding. The Australian orchid genus *Cryptostylis* is an excellent study subject because all five species attract the same male wasp pollinator despite obvious differences in flower shape and colour. Here I introduce this intriguing study system and present some initial observations of pollinator behaviour and interactions between pollinator learning and the dimensions of orchid patches.

Multimodal communication in the acoustic Lepidoptera: evolutionary pathways & biophysical constraints.

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Multimodal signaling, widespread among animal communications, may serve in conveying multiple messages, reaching different receiver categories, redundancy, and reliability. In Lepidoptera, sexual communication in various species involves male acoustic (ultrasonic) + chemical advertisement and female hearing + olfaction. These modalities operate during different stages of pair formation. Phylogenetic analysis indicates that olfaction is ancestral, with (male) chemical signaling and ultrasonic hearing (females, males) each evolving independently in multiple clades; hearing evolved as a response to predatory, echolocating bats. Finally, (male) acoustic signaling evolved in several clades possessing hearing. Dual use of the ultrasonic channel by acoustic Lepidoptera and bats indicates special problems for sexual communication in the former, as females must reliably distinguish male calls from bat echolocations. Moreover, they must distinguish these in flight and while perched, situations with exposure to different bats. Whereas multimodal communication may conceivably afford opportunities for adaptive response, studies of the pyralid moth *Achroia grisella* show that critical distinctions are made acoustically and via input from a third modality, proprioception: Females assess temporal characters of ultrasounds and respond appropriately in accordance with their assessment and situation, flying or in ground contact. This protocol may be particularly reliable, given the constraints of time and limitations of neural processing.

Reproductive strategies of cerambycid beetles & the role of semiochemicals.

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Reproductive strategies of adult cerambycid beetles, and the role of semiochemicals, are apparently associated with the condition of the host plant required by their larvae (whether hosts are alive, dying, or dead). For example, early studies have indicated that species whose larvae feed in living hosts are likely to use long-range pheromones, while those that feed in dying hosts typically rely on host plant volatiles to unite the sexes for mating. Nevertheless, more recent research has revealed evidence of long-range sex pheromones in dying-host species. There is also a growing body of evidence that mate recognition in species of both categories depends on contact chemoreception, and contact pheromones have been identified for several cerambycid species in recent years. Although the current state of cerambycid research documents considerable variation in reproductive strategy, the association with host plant condition provides structure for guiding future research.

Mass trapping of olive moths.

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A simple and inexpensive UV-trap was developed by the first author from materials generally available around the olive farm, to capture the main lepidopterous pests of olive trees. The trap consisted of a sticky-collecting device, two ultraviolet fluorescent tubes and tilted mirrors to increase light intensity by interference of reflected rays and spreading light reflection over a wider area. The trap was compared with delta-traps (baited with (Z)-Z-tetradecenol) and funnel traps (baited with (E)-11-hexadecenol and (E)-11-hexadecenyl-acetate) for monitoring the olive moth, *Prays oleae* and jasmine moth, *Palpita unionalis*, respectively. Sex pheromone catches (only males) were persistently lower in all collections of 2 successive olive seasons of 2002 & 2003 than those of UV-traps. *P. oleae* catches (males & females) in the UV-trap significantly ($P<0.01$) exceeded *P. unionalis* moths. The obtained results indicated that the UV-trap may represent an effective tool for mass-trapping of *P. oleae* and monitoring tool for *P. unionalis* and other species of moths in the area. The trap was so effective in trapping *P. oleae* even when adult population was low compared with sex pheromone trap. Some of non-target species may be protected from damage by placing a screen of proper sized mesh over the entrance of the trap. The UV-trap saved the fruit yield of a large olive farm without using any insecticides throughout the study period.

"New approaches to find key compounds in tritrophic interactions – exploiting genetic variation & *in situ* modification of volatile profiles. "

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Odour blends that are released by plants after herbivore attack can act as indirect defense by attracting natural enemies of the herbivore. These blends consist of mixtures of volatile organic compounds (VOCs), but, as yet, for many tritrophic systems it remains unknown, which VOCs of such complex blends are the most attractive ones. One way of studying the importance of individual VOCs is to test the attractiveness of odour blends differing in their pattern of compounds. This approach was used to test the attraction of naive and experienced females of the parasitoid wasp *Cotesia marginiventris* to volatile blends of maize infested with *Spodoptera littoralis* larvae. We used olfactometers in which female wasps were offered choices among odour blends of different chemical composition. The different blends were obtained by either using different maize inbred lines or by installing selectively adsorbing filters between the odour source and the olfactometer arms. The data were analysed with multivariate statistical techniques and revealed the relative importance of some VOCs that are often underestimated because of their small quantities.

How parasitoids read the news: information use in a patchy world.

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Patch leaving decisions in insect parasitoids have mainly been studied as a function of host density and host encounters in single-patch experiments. From such experiments we know that parasitoids respond to host kairomones and encounters with unparasitized hosts. Yet, parasitoids often encounter unparasitized as well as parasitized hosts. Response to encounters with parasitized hosts has been debated, but experimental evidence has been equivocal. Moreover, how information on a given patch and patch encounter rate influence the patch time allocation on subsequent patches has largely remained unanswered. Using *drosophila* parasitoids, we investigated behavioural responses of wasps to information cues for estimating patch quality and habitat quality. In patch-quality experiments we tested responses towards staged encounters with unparasitized vs. parasitized hosts. In habitat-quality experiments we tested whether odour from neighbouring patches affect the patch-time, and the searching behaviour when leaving the patch. Further, we studied how encounters with good and poor quality hosts and their density influence the time allocation on subsequent patches. Finally, we tested how parasitoids respond to variation in patch encounter rates in their patch time allocation and patch exploitation level. We discuss our results with respect of the reliability of such cues for future foraging success.

Floral scent mediating the foraging behaviour of some papilionid & danaid butterflies.

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In addition to visual stimuli, olfactory cues also play a significant role in the foraging behaviour of anthophilous butterflies. I investigated floral volatiles of *Citrus* flowers (white in colour) that stimulate flower-visiting by a papilionid butterfly, *Papilio protenor*, which shows innate colour preference for red, orange, and blue. To determine active compounds, odour components were first screened individually to assess their ability to evoke proboscis extension reflex (PER), and the actual attractancy of potential components was confirmed by free-flight experiments. Of 16 compounds identified from the flower, methyl anthranilate, 2-phenylethanol, and phenylacetonitrile were found most attractive to the butterfly, indicating that compounds with an aromatic ring serve as important olfactory cues in their flower-visiting. In Japan, flowers of *Eupatorium chinense* have been observed to strongly attract danaid butterflies in the genera *Parantica, Ideopsis, Euploea,* and *Salatura*. Although several volatile compounds detected from an isopentane extract of the flower showed weak to moderate PER-activities, none of them exhibited strong attractiveness to the danaids. However, floral nectar of the plant was found to contain a considerable amount of pyrrolizidine alkaloids. The major compound, identified as intermedine, strongly attracted some danaid species, suggesting that certain pyrrolizidine alkaloids *per se* function as attractants.

Molecular cloning of a putative PBAN receptor from pheromone glands of the silkmoth, *Bombyx mori*.

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Despite recent successes in elucidating the molecular components of sex pheromone biosynthetic pathways within the Lepidoptera, identification of the gatekeeper of the pathway, the PBAN receptor, has proven to be elusive. Given the sequence similarity between the active C-terminal regions of PBAN and neuromedin U, it was hypothesized that their respective receptors might also be similar in structure. Consequently, a homology-based molecular approach utilizing primers constructed from the conserved regions of insect neuromedin U receptor homologues was used to amplify a full-length 2777-nt clone from a *Bombyx mori* pheromone gland cDNA library. Conceptual translation of the ORF indicated that it encoded a 45.8 kDa 7 transmembrane protein with 82% identity to a GPCR that has been recently cloned from pheromone glands of *Helicoverpa zea*. Interestingly, the sequence identity decreases considerably within the amino terminus and the *Bombyx* receptor contains a 67 amino acid C-terminal extension. Tissue distribution analyses reveal that the receptor is specific to the pheromone gland where it undergoes significant up-regulation in the day preceding eclosion. In addition, when expressed in Sf9 cells the receptor not only exhibits specific binding to PBAN but also mobilizes calcium in a dose-dependent response.

Reception of floral odours by *Helicoverpa armigera* antennae.

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The antennal receptor cell complex of *Helicoverpa armigera*, that is used in detecting a range of floral and green leaf odours, was investigated with electrophysiological and behavioural methods. By using mixtures of odours at saturating levels, the electroantennogram method can produce useful information about the classes of receptor cells used in detecting plant odours. This methodology provides information that both differs from and augments that gained by single neuron recordings, and as such allows the testing of hypotheses related to receptor types. A range of volatiles was tested including those principally associated with flowering parts or leaves as well as volatiles common to both. The two enantiomers of the chemical μ-pinene were also investigated and results suggest a common receptor neuron type for these enantiomers. Behavioural experiments showed that conditioned learning of odours can influence the decisions of the insect.

Significance of male's licking female dorsum in matings of Cerambycidae (Coleoptera).

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In matings of, if not all, some coleopteran families including Cerambycidae, it has been often recognized that male, on female's back, lick her dorsum with his mouth parts. However, there has been only a very limited number of literature on this behavioral component. We therefore investigated the significance of each of the body parts in cerambycid mating sequence, focusing on males' dorsum-licking behavior. Adults of diurnal cerambycid species, *Dinoptera minuta* (Gebler) (Lepturinae) and *Chlorophorus quinquefasciatus* (Cerambycinae), were collected in the field, and each of the test animals was treated by mutilating antennae with scissors (male, female) or coating its compound eyes (male, female) or pronotum plus scutellum (female) with water-based white correcting fluid. A treated individual and an untreated partner were paired and their mating sequence followed. Untreated pairs initiated mounting directly upon male antenna's touching female's body or antenna. Antenna-mutilation of male caused severe hinance of mating. Blind males did not mount or copulate. Coating pronotum/scutellum-region of the female also caused copulation hinance. The last result suggests that female beetle possibly possesses in her pronotum/scutellum region a source of pheromone toward the male and/or a sensory organ admitting male's appeasement by his mouth parts while in copula.

Using synthetic herbivore-induced plant volatiles to enhance conservation biological control in crop ecosystems: field experiments in hops & grapes.

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The potential of using synthetic herbivore-induced plant volatiles to enhance conservation biological control of insects and mites in crop ecosystems is being researched in hops and grapes in Washington State. Results to date indicate that a number of natural enemy species in the families, Chrysopidae, Hemerobiidae, Anthocoridae, Geocoridae, Miridae, Coccinellidae, Syrphidae, Braconidae, Empididae and Mymaridae, are attracted to sticky traps baited with aqueous methyl salicylate (MeSA), hexenyl acetate, farnesene or octyl aldehyde. Further, hop yards and grape blocks baited with slow release sachet dispensers of MeSA appeared to recruit larger populations of some insect predators (e.g. *Stethorus punctum picipes, Orius tristicolor, Chrysopa nigricornis*) than unbaited areas, resulting in some instances, in improved biological control of spider mites. The use of synthetic plant-signalling compounds as 'Herbivore-Induced Plant Protection Odors' (HIPPOs) may provide a novel yet practical option for improving the efficacy and reliability of conservation biological control in a variety of agricultural ecosystems.

Mating behaviour of the New Zealand native leafroller *Cnephasia jactatana* Walker (Lepidoptera: Tortricidae).

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Cnephasia jactatana Walker is a pest of kiwifruit in New Zealand. The objective of this study was to provide information on its mating behaviour for the development of pest management measures. Sexual maturation period was 3 d for females and 2 d for males. More than 50% of eggs were laid within 3 d after mating. Female reproductive fitness was reduced by > 85% if mating was delayed for 4 d. Males actively approached and courted females before mating occurred and females appeared to be less active. Males used their antennae, labial palps and wings to touch females when courting. Virgin males performed more mating attempts than mated males when competing for females. Sexual selection in *C. jactatana* did not support Darwin's sexual selection theory where females are more selective and males suffer the sexual selection effects. In this species, males appeared to be more selective than females. Both sexes preferred virgin to mated mates for mating. Males preferred young to old females for mating while females did not discriminate the male age when choosing a mate. Male body weight did not confer any mating advantage. However, females preferred males with longer antennae for mating. Light- and average-weight males presented a size assortive-selection while heavy males did not show any preference.

Regulation of pheromone biosynthesis in moths: PBAN & a G-protein coupled receptor.

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Pheromone biosynthesis in most moths is regulated by PBAN (pheromone biosynthesis activating neuropeptide). This hormone is produced by the subesophageal ganglion and can be released into the hemolymph to stimulate biosynthesis in pheromone glands. PBAN acts directly on pheromone glands using cAMP and calcium as second messengers. Biosynthesis is activated by stimulating acetyl-CoA carboxylase or a reductase. Pharmacological evidence indicated the presence of a G-protein coupled receptor. To identify the receptor we utilized the finding that *Drosophila* contains PBAN-like peptides and thus a receptor for the peptides. Three *Drosophila* receptors had sequence homology to neuromedin U receptors from vertebrates. Neuromedin U will stimulate pheromone biosynthesis in *Helicoverpa zea* and *H. armigera* indicating cross-reactivity of the peptide and functional homology of the receptors. Based on sequence homology to the receptors from *Drosophila,* the open reading frame of a gene was isolated from mRNA obtained from pheromone glands of *H. zea.* The PBAN-receptor was cloned and expressed in Sf9 cells and using a calcium mobilization assay responded to PBAN in a dose-dependent manner. Additional peptides produced by the PBAN family were also tested. This represents the first identification of a receptor for the PBAN family of peptides with a known function in female moths.

Herbivore-induced volatiles alter plant resistance.

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In response to damage, several plants have been found to emit volatiles that make neighboring plants more resistant to herbivory. When sagebrush is damaged in the field, airborne volatiles cause neighboring conspecifics to change chemically and suffer less damage by naturally occurring herbivores. Emissions of methyl jasmonate increase in the headspace of damaged sagebrush, making it a leading candidate as a volatile cue mediating increased resistance. Since many plant species respond to methyl jasmonate, it seemed plausible that various neighbors might induce resistance when growing near damaged sagebrush. Damage to sagebrush in the field caused neighboring wild tobacco but not other naturally occurring plant species to become more resistant. Preliminary laboratory experiments suggest that methyl jasmonate is sufficient, but not necessary, as an airborne cue.

Effective pheromone lures for monitoring the rice leaffolder moth, *Cnaphalocrocis medinalis* (Lepidoptera: Crambidae).

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Several factors affecting the attractiveness of traps baited with synthetic sex pheromone components of *Cnaphalocrocis medinalis* were examined in the laboratory and field. In the laboratory, among the four pheromone components, Z11-18:Ald, Z13-18:Ald, Z11-18:OH and Z13-18:OH, the major component, Z13-18:Ald, was shown to be an essential component for attraction and Z11-18:Ald was shown to be a potent synergist for the major component. The alcohols also showed an apparent synergistic effect in the laboratory bioassay when their mixture was combined with an aldehyde mixture. The addition of the geometric isomers of the four components at levels greater than 5% of the purified 4-component blend showed significant inhibitory effects on the pheromonal activity in the laboratory. In the field, no synergistic effect of the alcohols on the aldehydes was shown. A range of loadings of the pheromone rubber septa (0.01–3.42 mg) was tested and all remained attractive for at least one month with optimum attractiveness on 0.86 mg/septum. No significant effects were obtained when the ratio of Z11-18:Ald to Z13-18:Ald was changed. Rubber septa containing 0.86 mg of the four components (*E*-isomers 5%) at the natural ratio are being effectively used for monitoring.

Experimental evolution of improved learning ability in *Drosophila.*

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While fitness benefits of animal learning are well understood, other questions concerning the evolution of learning remain open. How easily can improved learning ability evolve? Does evolution act on "general intelligence", or rather on learning abilities specialized to particular tasks and situations? What are the costs of learning ability? To address these questions we have exposed replicated populations of *Drosophila melanogaster* to long-term selection for improved olfactory-gustatory learning in the context of oviposition substrate choice. After over 20 generations of selection the experimental populations evolved markedly improved learning ability. The improved learning ability generalized to other tasks, which involved different stimuli, reward rather than aversion learning, and classic rather than operant conditioning. The selected populations also showed better learning ability as larvae. This indicates that the response to selection was based on evolutionary changes in general aspects of the learning and memory, involved in many forms of learning. Furthermore, compared to control populations, larvae of the selected populations showed lower competitive ability. Additional data excluded inbreeding as a potential cause of reduced competitive ability. Therefore we believe that the lower competitive ability of the experimental populations indicates a genetic cost of learning ability.

Patch leaving decisions of *Cotesia rubecula*: a synthesis.

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Cotesia rubecula (Hymenoptera: Braconidae) is a solitary endoparasitoid of larval *Pieris rapae* (Lepidoptera: Pieridae). Adult P. *rapae* lay single eggs on a range of *Brassica* species (Brassicaceae) and other plants, and their larvae develop as individuals on host plants. *C. rubecula* parasitises first through fourth instars of *P. rapae*. A series of laboratory and field experiments has examined patch-leaving decisions of Australian *C. rubecula*. A range of factors has been shown to influence patch-leaving tendency of this parasitoid including host stage and density, the amount of plant damage by the host, recent oviposition experience of the wasp and the wasp's feeding status. Decisions are also influenced by the spatial distribution of host plants. This paper will develop a synthesis of experimental results and aims to identify the selective factors that have shaped the foraging behaviour of *C. rubecula*.

The infrared organ of the Australian “little ash beetle” *Acanthocnemus nigricans*: morphology of the specialised sensilla.

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The Australian beetle *Acanthocnemus nigricans* (Coleoptera, Acanthocnemidae) approaches forest fires. The reason for this pyrophilous behaviour might be that larval development depends on burned wood. *A. nigricans* has a pair of infrared organs on the ventrolateral sides of its prothorax. Each organ consists of a cuticular “sensory disc”with a diameter of about 150µm. About 70 small cuticular sensilla were found mainly on the outer surface of the disc. The cone shaped cuticular peg of a sensillum is about 2µm in diameter. It has no pores and is surrounded by a cuticular wall. The single sensory cell of the sensillum shows some similarities to the “bauplan” of a mechanoreceptive sensory cell of a trichoid sensillum. However, three specialisations can be found: (i) the denitic outer segment is enveloped by a pronounced electron-dense structure, which most likely represents the hypertrophied denitic sheath. (ii) The membranes of the denitic inner segment and the soma show distinct infoldings which contain a high amount of mitochondria. (iii) No tubular body can be found.As the cuticular disc responds to infrared radiation, it is likely that this specialised new type of insect sensillum may act as a thermoreceptor. Further examinations will have to test this hypothesis.

Pheromone response & mating activity in the consperse stink bug, *Euschistus conspersus.*

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The consperse stink bug, *Euschistus conspersus* Uhler, has emerged as an important pest of pome fruit production in Washington state. My research has focused upon quantifying the behavioural response of this insect to its aggregation pheromone, methyl (2*E*,4*Z*)-decadienoate in an attempt to develop pheromone-based monitoring or management systems. I describe research investigating the pheromone-mediated aggregation behaviour in *E. conspersus* in the field and describe the subsequent development of a detailed diel activity profile for this insect. *E. conspersus* aggregations peaked at dusk, with mating occurring primarily at night. Sex ratios were equal in aggregations and both sexes were found to be polygamous, mating on successive nights but with different partners. Laboratory experiments were performed to investigate possible factors mitigating mate choice in this species. The significance of these findings with respect to the possible function of aggregation in this species is discussed, as well as the implications for management.

Molecular basis of pheromone binding, release, & inactivation.

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Insects perceive the world through small molecules - pheromones and other semiochemicals. Their odor-oriented navigation relies heavily on the ability of the olfactory system to selectively detect minute amounts of pheromones and reset the detectors in millisecond scale (by inactivating the chemical signals). Pheromone molecules enter the sensillar lymph through pore tubules in the sensillar wall, are solubilized upon being encapsulated by odorant-binding proteins (OBPs), and transported to the olfactory receptors (ORs). Upon interaction with negatively-charged sites at the denitic membrane, the OBP-ligand complex undergoes a conformational change that leads to the ejection of pheromone to activate the pheromone receptors. While bound to OBPs, pheromone molecules are protected from odorant-degrading enzymes (ODEs). In this presentation, we will (1) briefly discuss the molecular mechanisms of pheromone binding and release by focusing on the *Bombyx mori* system, i.e., how bombykol enters and leaves the binding cavity in BmPBP, (2) report the first odorant-degrading enzymes isolated from insect antennae to date, and (3) discuss the possible mechanism of enzymatic inactivation of pheromones.

Induction of preference for oviposition repellents originated from non-host plants by a specialist herbivore.

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Repellents derived from non-host plants have been assumed to have a role to play in the behavioural manipulation of crop insect pests. However, behavioural responses to such repellents by insects have been rarely observed either in the laboratory or field. In laboratory and greenhouse experiments, we observed the responses of diamondback moth, *Plutella xylostella*, a special herbivore of crucifer plants to oviposition repellents derived from a non-host plant, *Chrysanthemum morifolium.* When provided with a choice between untreated Chinese cabbage and Chinese cabbage sprayed with an extract of *C. morifolium* in one arena, inexperienced females deposited 9-fold more eggs on the former than on the latter plant, demonstrating that the plant extract had substantial repellent effects on this insect. However, when the test adults were offered prior experiences of the plant extract and then exposed to the dual-choice of plants for oviposition, they laid significantly more eggs on the plant sprayed with the *C. morifolium* extract than that on the untreated plant. Further observations with neem-based materials also showed induction preference to these repellents by *P. xylostella.* Adult learning thus can render repellents derived from non-host plants to become ineffective or even attractive to this insect.

Non-host antifeedants in hylobius pine weevils: from lab to field applications.

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We searched for antifeedant activity in non-host woody plants to find new compounds for seedling protection of Scots pine (*Pinus sylvestris*) and Norway spruce (*Picea abies*) against pine weevil feeding. In total, 38 species from 25 families were compared. The weevils removed less of both outer and inner bark in *Ilex*, *Evonymus*, *Populus*, *Syringa*, *Taxus*, *Tilia*, *Viburnum*, *Lonicera*, and *Sorbus* than on *Pinus*. In some plants, *Juglans*, *Fraxinus*, *Sambucus*, *Aesculus*, *Quercus*, *Corylus*, *Fagus*, *Salix*, *Alnus*, & *Acer*, the outer bark was removed by the insects but had an inner bark with antifeedant qualities. Bark of Linden, *Tilia cordata*, showed the best antifeedant qualities and was extracted and tested for antifeedant activity. The active extract was fractioned on silica gel. TLC and GC was used to trace chemical similarities between the fractions and resulted in merging the number of fractions down. The micro assay gave positive antifeedant activity in two adjacent fractions. These fractions were analyzed and nonanoic acid was found in both of them. Synthetic octanoic, nonanoic, and decanoic acid tested in the micro feeding assay had strong antifeedant activities against both sexes.

Characterization of genes encoding enzymes involved in bombykol biosynthesis.

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In the biosynthesis of class I-type Lepidopteran pheromone components, the key enzyme required to produce the oxygenated functional groups is pgFAR. This enzyme converts fatty-acyl pheromone precursors to their corresponding alcohols, which can then be, depending on the moth species, further modified via acetylation or oxidation reactions. In experiments designed to characterize the pgFAR in *B. mori*, we have isolated a cDNA clone encoding a protein homologous to a FAR from the desert shrub, *Simmondsia chinensis*, commonly known as jojoba. The deduced amino acid sequence of this clone predicts a 460 amino acid protein with a consensus NAD(P)H binding motif within the amino terminus. Northern blot analyses indicated that the 2-kb transcripts of this gene are specifically expressed in the pheromone gland at one day prior to adult eclosion. Functional expression of this gene in the yeast *Saccharomyces cerevisiae* not only confirmed the long-chain FAR activity but also indicated a distinct substrate specificity. Finally, the transformed yeast cells evoked typical mating behavior in male moths when cultured with the pheromone precursor fatty acid. We will also refer to a bifunctional desaturase utilized in the production of the conjugated diene system from which bombykol biosynthesis is dependent on.

"Come on baby light a fire": the reproductive biology of *Actebia fennica.*

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Black army cutworm out breaks are associate recently burned areas, which are unpredictable in time and space. Our wind tunnel experiments showed that while females exhibit oriented, upwind flight in smoke plumes males do not. We also examined the effect of smoke volatiles the temporal changes in juvenile hormone biosynthesis and oocyte development, as well as on the age at which females first exhibited the behaviour associated with the emission of the sex pheromone. All data support the hypothesis that volatiles from burnt vegetation serve as " kairomones", accelerating sexual maturation in females. However, mated females, held in the presence of volatiles that stimulated sexual maturation, actually laid fewer eggs. An ecological hand-waving scenario will be proposed to explain the results obtained to date. We will also discuss how the recent identification of different smoke compounds detected by females might subsequently be used in management programmes for this pest species.

On the kairomonal attraction of adults & nymphs of *Elatophilus* spp. (Heteroptea: Anthocoridae) to sex pheromones of *Matsucoccus* spp. (Hemiptera: Matsucoccidae).

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While kairomonal response of insect predators to the sex pheromone of the insect prey has been rarely documented, our recent studies proved consistently that the female sex pheromones of *Matsucoccus josephi* , *M. feytaudi* and *M. matsumurae* are powerful long range attractants for *Elatophilus* spp. Examining the attraction to the pheromone and analogs of seven *Elatophilus* spp. revealed a clear species specific attraction displaying by these predators. Similarly to their conspecific adults the nymphs of *E. hebraicus, E. crassicornis* and *E. nigricornis* are also lured by the specific sex pheromone. It seems that sex pheromones of *Matsucoccus* serve *Elatophilus* spp. as aggregation kairomones and as sexual stimulants. *Elatophilus* spp. display courtship behavior and mate frequently in arenas baited with the pheromone. When the predator population is high, most of the females that congregated around a pheromone source were already mated but displaying none or few visible eggs in the ovaries. We suggest that the typical group movement displayed by the nymphs is determined by their attraction to the pheromone. This behavior may increase survival through "a better feed tracing" and "safety in numbers". Female first mating probably occur soon after molting within their own group before being lured to a pheromone source.

Odor perception & odor learning in honeybees.

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Honeybees discriminate a large range of odors, and learn odors as signals for food. We have used optical imaging and intracellular recording techniques to examine the neural substrates of odor coding and memory formation in the antennal lobe (AL) and the mushroom bodies (MB). Odors elicit combinatorial patterns of activity in the glomeruli of the AL. Imaging of the input region of the mushroom bodies reveals that odors are represented in specific patterns of microglomerular activation. A comparison of the dynamics of the odor responses shows that mushroom body intrinsic neurons code odors in a sparse way with a small number of spikes per stimulus and a small number on activated Kenyon cells. Odor learning leads to an enhancement of the synaptic transmission at the input synapses to the Kenyon cells that respond to the trained odor (CS+), and a reduction of synaptic efficiency to the non trained odor (CS-). A model will be presented that captures the results of our electro- and optophysiological recordings, and assigns particular functions to the mushroom body in the bee brain.

Male-produced aggregation pheromone of *Neoclytus acuminatus acuminatus*.

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The red-headed ash borer, *Neoclytus acuminatus acuminatus* (F.) (Coleoptera: Cerambycidae) is endemic to North America, where it infests a variety of hardwood trees. It also has been accidentally introduced into Europe and possibly New Zealand. In the field, we observed males performing a stereotyped calling behavior which provided the first indication of pheromone production by male *N. a. acuminatus*. This behavior has not been reported for any other cerambycid species. Insects of both sexes were attracted to caged males but not to females. We report the identification and field testing of a male-produced aggregation pheromone for this species. During field trials, females of another cerambycid species, *Curius dentatus* Newman, were attracted to traps baited with some of the test compounds, suggesting that one of more of the test compounds were also part of the sex pheromone of this relatively unknown species as well. These may be the first examples of cerambycid pheromones with an activity range of tens to a huned meters or more.

Pheromonal & vibrational signals of stink bugs in the genus *Chlorochroa*.

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Stink bugs in the genus *Chlorochroa*, similar to other phytophagous stink bugs, use a combination of pheromonal and substrate-borne vibrational signals in their reproductive behaviors. The pheromones are male-produced, and probably act as attractants over longer distances, whereas vibrational signals (songs) are produced by both sexes once the bugs are on the same plant. The vibrational signals are sex- and species specific, with males and females producing a repertoire of calling songs as orientation cues, and courtship songs once the bugs are close together or in contact. The pheromonal and vibrational signals will be compared and contrasted for three sympatric *Chlorochroa* species, and the roles of these signals in intraspecific communication will be discussed.

Fruitspotting bugs.

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The fruitspotting bugs, *Amblypelta nitida*, and the closely-related *Amblypelta lutescens*, are serious pests of tropical and sub-tropical fruit and nut crops in Queensland and northern New South Wales. Current control relies on the frequent application of insecticides, with recent investigations focussing on promising soft-option strategies such as restricting insecticide application to fruitspotting bug hotspots, and the use of SurroundÒ to deter feeding. Behaviour-modification approaches are a legitimate component of clean-green management strategies, and to this end, investigations of the insects' semiochemistry were commenced 14 years ago. Despite early breakthroughs in the chemistry studies, subsequent progress has been sporadic. The major component of male-derived volatiles of *A. nitida* was shown to be a mono-epoxide of (E)-b-ocimene, having the trivial name, myroxyde; and of *A. lutescens*, (-)-(3R)-(E)-nerolidol. The absolute configuration of the former compound has been defined through the development of efficient synthetic strategies and the application of SPME sampling technology. Recent gas chromatography-electroantennography studies have confirmed myroxide, along with the minor compounds, nonanal and decanal, as stimulants of antennal response in *A. nitida*. The attractiveness of blends of racemic myroxyde and the aldehydes, using a new clear-plastic football-shaped trap, is being investigated.

Isolation & characterisation of a putative aggregation pheromone component for the striped cucumber beetle, *Acalymma vittatum* (F) (Coleoptera: Chrysomelidae).

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Male-produced aggregation pheromones have previously been identified from a limited number of Chrysomelids; the hydroxy-ketones (*S*)-3,7-dimethyl-2-oxo-6-octene-1,3-diol from the Colorado potato beetle, *Leptinotarsa decemlineata* (Say) [1], and (*E*)-8-hydroxy-6-methyl-6-octen-3-one from the cereal leaf beetle, *Oulema melanopus* L. [2]. Himachalene and cadinene sesquiterpenes from *Phyllotreta cruciferae* and three *Apthona* spp. of flea beetles were electrophysiologically active [3]. Field trapping studies showed that feeding male *A. vittatum* attracted significantly more congeners than feeding females, or undamaged cucumber plants, indicating that it may also use an aggregation pheromone [4]. GC-EAD studies were carried out using airborne collections from feeding beetles, or SPME collections, and male or female antennae. One component of male collections consistently gave a strong EAD response, and was absent from collections from female beetles. This putative pheromone was initially characterised by GC-MS and micro-reactions. More than 3000 beetle-days of collection on Porapak resulted in sufficient compound for extensive 1- and 2-D NMR spectroscopic studies, and FTIR. These allowed complete structural characterisation of the pheromone. The only closely related compounds are antibiotics of microbial origin, which were compared with the pheromone by NMR. Currently, efforts to synthesize the pheromone, and test for behavioural activity are in progress.

Mate location & mating behaviour of ***Chrysophtharta agricola*** **(Chapuis) (Coleoptera: Chrysomelidae: Paropsini).**

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The mating system of *Chrysophtharta agricola* comprises prolonged searching scramble competition polyandry, whereby receptive females are evenly dispersed spatially and temporally, and males search competitively for them: 90 % of beetles captured on sticky traps were males. More males were caught on traps baited with sexually receptive conspecific beetles than on traps that contained foliage only. Male mating behaviour comprised long pairing associations with several intromission events, and did not change according to female mated status. Paternity analysis using a polymorphic gene for melanism showed various sperm use over lifetime offspring production. Field data were modelled to estimate that beetles mate 15 – 51 times in a season. Beetles did not select mates based on size, colour, generation or parasitic mite infection.

Sex pheromone of a carpenter moth, *Cossus insularis* **(Staudinger).**

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Cossus insularis (Staudinger) (Cossidae: Lepidoptera) is a woodborer found in Honshu, Kyushu and Tsushima Island of Japan. The larvae excavate their galleries in a group on twigs or trunks of the broad-leaved trees such as willow trees, pear trees, chestnut trees, and cause damage and often mortality of the trees. To utilize sex pheromone for monitoring or controlling the borers, sex pheromone of *C. insularis* was identified. Combined gas chromatography-electroantennographic detection (GC-EAD) analysis of airborne volatiles collected from unmated female moths by a solid-phase microextraction (SPME) showed that two volatile compounds elicited significant electroantennographic responses from the antennae of male moths. These two volatile compounds collected from female moths were identified as (*E*)-3-tetradecenyl acetate and (*Z*)-3-tetradecenyl acetate by mass spectral and retention index comparisons with synthetic standards. In field bioassays, sticky traps baited with a lure of a 4:1 blend of synthetic (*E*)-3-tetradecenyl acetate and (*Z*)-3-tetradecenyl acetate captured significantly large number of male moths, while unbaited traps captured no male moths. These results indicate that sex pheromone of *C. insularis* is a mixture of (*E*)-3-tetradecenyl acetate and (*Z*)-3-tetradecenyl acetate.

Plant/plant communication-herbivores & their natural enemies: experiments with spring-sown barley.

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Experiments with a barley crop and weeds have shown that effects of chemical communication between plants can be traced in a tritrophic perspective as follows:*Exposure to allelobiotic substances has a physiological effect on cereal plants, evidenced by significant changes in the pattern of biomass allocation and changes in leaf temperature. *Exposing cereal plants to allelobiotic volatiles from neighbouring plants and root exudates from aggressive weeds decreases the acceptability of the exposed plants for aphids. This happens in interactions between different cultivars of barley (*intra-species*), and between cereals and a number of aggressive weeds (*inter-species*). *Exposure to allelobiosis from weeds causes barley plants to become more attractive to aphid natural enemies, a ladybird *Coccinella septempunctata.* *The results suggest that plant responses to allelobiosis is a preparation for coexistence with other plants and this is a basis for follow up effects in higher trophic levels. The positive effect on searching behaviour of polyphagous predators may increase the ecological success of "listening" plants in a mixed plant stand and, also have negative effects on herbivore plant acceptance.

Artificial substrate bioassay for testing oviposition of the southern green stink bug, *Nezara viridula* (L.) conditioned by soybean plant chemical extracts.

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The southern green stink bug, *Nezara viridula* (L.) (Heteroptera: Pentatomidae) is effectively controlled with egg parasitoids, and egg production is an important component of this strategy. Therefore, a laboratory bioassay was developed for testing its oviposition preference toward chemicals extracted from soybean pods and leaves. An artificial substrate (cheesecloth) was stretched over a wooden ring (embroidery hoops), treated with plant extracts or chromatographic fractions, and then exposed to adult stink bugs to assess oviposition preference. Freeze-ied soybean leaves and pods were extracted with hexane, then with methanol, and finally with water. The organic solvent extracts were fractionated by liquid chromatography, on a C_{18} reverse phase column, on a normal phase silica column, and by thin layer chromatography (TLC) on silica gel. The methanol extract of pods showed the greatest bioassay activity. The most active reverse phase fraction derived from this extract was eluted with 20% methanol in water. After subjecting this fraction to chromatography, the greatest activity occurred in the fraction eluted with 60% methanol in methylene chloride. TLC gave no single fraction with demonstrated activity, but the recombined fractions were again active, indicating that multiple components elicit oviposition. Hairs (long and short) on the female genitalia might be involved in chemical and/or mechanical recognition of oviposition stimulants.

The chemistry of sexual deception in Australian orchids.

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Pollination by sexual deception is among the most intriguing of orchid pollination syndromes. Odors are well established as the primary stimuli for sexually attracting the male insect pollinators in these orchids, but we have only begun to discover how many, and which compounds are involved. Using gas chromatography with electroantennographic detection (GC-EAD), GC-mass spectrometry (GC-MS) and GC-IR-spectrometry we have determined that a single active compound, Chiloglottone, produced by both females of the wasp *Neozeloboria cryptoides* and flowers of the orchid *Chiloglottis trapeziformis* is active in antennal neurons of the pollinators, males of *N. cryptoides*. This compound, tested as synthetic copy, proved to be equally attractive to natural extracts of flowers and wasps. We are now determining the chemical identity of sexual attractants in other *Chiloglottis* orchids. GC-EAD and GC-MS analysis indicates only 1 or 2 active compounds are involved and these compounds are related to Chiloglottone. Pollinator specificity and associated reproductive isolation is therefore likely under simple genetic control in these orchids. Thus rapid sympatric speciation is possible. We will present an update of our multifaceted research program that is exploring this hypothesis of sympatric speciation.

Biochemistry & evolution of floral scent.

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Floral scents of plants are often complex mixtures of volatiles derived from several metabolic pathways. The number of presently known volatile compounds from flowers is approaching a thousand, and new compounds are constantly being discovered in various plant species. Closely related species have distinct volatile profiles that can be discriminated by specialized insect pollinators, thus limiting gene flow across species boundaries. The ability of related plant species to synthesize different volatiles in their flowers is sometimes due to differential gene expression. However, new enzymes that catalyze the formation of new floral scent components are constantly evolving through the process of gene duplication and divergence, allowing for the majority of floral scent divergence. It has been shown that very small changes in protein sequences can lead to the ability of the changed enzymes to catalyze the transformation of drastically different substrates, leading to the production of new floral scent compounds. Such evolution is facilitated by the presence of large gene families in plant genomes that already encode similar enzymes with slightly different substrate specificities; the large number of such genes makes genes duplications even more likely to occur.

Herbivore-induced plant volatiles identified from interactions at higher trophic levels: persistent effects on plants in the laboratory & field.

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In many cases, volatiles produced by plant defensive mechanisms during herbivory act directly to repel insect attack, and may have indirect effects by attracting predators and parasitoids of the pest insect. *cis*-Jasmone occurs as a component of flower volatiles, but can also be produced by damaged vegetative tissues. In an olfactometer bioassay, *cis*-jasmone was repellent to alatae of the grain aphid, *Sitobion avenae* (Homoptera: Aphididae). *cis*-Jasmone is a catabolite of the stress-produced jasmonic acid, which can induce plant defence, but had previously been considered only as a biological sink for the jasmonate pathway. However, bean plants, *Vicia faba*, exposed to *cis*-jasmone vapour were more attractive to the parasitoid *Aphidius ervi*. Moreover, wheat, *Triticum aestivum*, seedlings sprayed with formulated *cis*-jasmone 24 h previously were less susceptible to attack by *S. avenae* than control plants. In field simulator studies, significantly fewer alate *S. avenae* settled on *cis*-jasmone treated plants over a 24 h period. The intrinsic rate of population increase, r_m, of *S. avenae* apterae was reduced on *cis*-jasmone treated seedlings. In a series of small-plot experiments conducted over four years, *cis*-jasmone applications reduced cereal aphid populations infesting wheat in the field.

Polyphagy in *Helicoverpa armigera*: primary host plants, host-associated volatiles & neurosensory stimulation.

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Headspace volatiles of a subset of known host plants and non host plants of *Helicoverpa armigera* were screened by the use of Gas Chromatography linked with Electroantennography. Pigeon pea (*Cajanus cajan* L.) is one of the most attractive hosts to *H. armigera* in India, so was included as the primary reference plant to compare the presence and concentrations of electrophysiologically active chemicals across the test plants. Eight active compounds were identified in pigeon pea headspace volatiles in relatively higher concentrations. They include three green leaf volatiles ((E)-2-hexenal, Z-3-hexenylacetate and Z-3-hexenyl-2-methylbutyrate) and five monoterpenes (α-pinene, β-myrcene, Limonene (E) – β-ocimene, and linalool). The other known host plants have a small subset of these chemicals, whereas the non host plants contained even fewer. Green leaf volatiles were prominent in the host plants. The Australian *H. armigera* individuals used in this study did not respond to the group of sesquiterpenes present in pigeon pea headspace, and thus differ from their Indian counterparts. The physiological status and the sex of the moths had no effect on their responses to these chemicals. The study exemplifies how some host plants species are primary targets for searching moths, whereas the other host plants are likely to be incidental targets.

Experimental evidence of semiochemicals modulating the behavior of *Loxosceles intermedia* Mello-Leitão, 1934 (Araneae: Sicariidae).

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There is evidence of semiochemicals modulating the behavior of many species of spiders, but few reports regarding the isolation, chemical identification, and synthesis of semiochemicals in these arthropods. Our search is experimentally focused on the occurrence of volatile semiochemicals modulating the behavior *of L. intermedia*, a venomous spider that constitutes a serious public health problem in Curitiba, Brazil (3,334 people bitten in 2002). An olfactometer of rectangular arena was used. The individuals employed as stimuli were put in metallic cages, positioned against air flow. We tested all possible combinations of males and females as stimuli or test individuals. For each stimuli trial different individuals of *L. intermedia* were used. Tests were made on summer and winter 2003, and summer 2004. Only females tested during summer significantly attracted males, indicating a volatile pheromone produced during the summer season. Pheromone glands are not described for spiders, but we suppose the secretory cells are less active during winter. This is the first evidence of semiochemical in the *Loxosceles* genus. Studies involving the extraction and GC-MS identification of the putative semiochemical(s), as well as further experimental observations are being carried out.

Evolution of pheromone biosynthesis in moths.

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In addition to their role in metabolism, acyl-CoA desaturases in moths play a key role in female sex-pheromone biosynthetic pathways. These atypical membrane-bound sex pheromone desaturases have evolved a wide range of regio- and stereospecificities. Recently, it was shown that changes in pheromone blends in moths of the genes *Ostrinia* result from unusual molecular mechanisms that influence the expression and evolution of sex pheromone desaturases. In this talk, an overview of sex pheromone desaturase evolution is presented that covers the phylogeny and evolutionary patterns of sex pheromone genes, the moth sex pheromone molecular clock, and genomics of sex pheromones.

Dynamic irritability in parasitoids.

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Parasitoids may encounter other organisms while foraging on host-containing patches. Some of those organisms post direct threats to the foraging wasp. On the other hand, the patch provides resources that that can directly contribute to the parasitoid's reproductive success. Thus, the parasitoids must "decide" how to tradeoff threats versus benefits. Further complicating matters are several parameters that can affect costs and benefits, including: (1) the threatening organism may or may not attack, (2) the patch will diminish in value over time and (3) forager state, in particular, egg load, body mass and energy state. For example, heavier wasps may be less agile or slower in escape than light individuals. Here I consider 2 mutually exclusive decisions, continue foraging vs. abandon the patch as a function of the three aforementioned parameters. In my talk I will develop state-dependent theory and test some predictions from the theory as to conditions under which parasitoids will abandon patches following encounters with threats. I will show both theoretically and experimentally that sensitivity to threats are dynamic i.e. they depend upon parasitoid internal state and perception of danger.

Herbivore-induced volatiles: variability in production & variability in natural enemy response.

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Ecological interactions between plants and the enemies of their enemies are often mediated by information available as blends of volatile chemical compounds. Because the information is under control of the sender and the receiver cannot check its validity from a distance, the receiver has to base its decisions on previous experiences and this creates opportunity for the sender to cheat. Hence, we expect conflicts of interest between the sender and the receiver. We will present recent evidence for plants that cheat, i.e. produce amounts of chemical information that does not reflect the number of herbivores, so that the predators fail to select plants harbouring the highest densities of their prey. We will also present evidence showing that predatory arthropods can only partly cope with this plant-generated variability by associative learning. Based on theoretical models of the evolution of chemical languages and classic predator-herbivore-plant models extended with an information-perception interphase, we predict that the emergent non-linearities in plant-predator communication have dramatic consequences for the evolutionary and ecological dynamics of tritrophic interactions.

Influence of temperature on mechanosensory host location by vibrational sounding in parasitic wasps.

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Although temperature is one of the most important environmental parameters, there are relatively few studies dealing with its influence on ecological processes between trophic levels, e.g. on host-location mechanisms of parasitoids. Certain parasitic wasps use self-produced vibrations transmitted on plant parts to locate their immobile hosts (lepidopteran pupae) by resonance signals. This mechanosensory mechanism, vibrational sounding, depends both on physical cues of the environment and physical activity of the parasitoid and, therefore, should depend on ambient temperature and the likely adaptation of the insects to their environmental temperature range. We analysed such influences by choice experiments with plant-stem models in the temperate ichneumonid *Pimpla turionellae* and the tropical relative *Xanthopimpla stemmator*. Our results show a substantial effect of temperature on host-location activity and on the success of the process. Outside optimum ranges, the performance of the wasps decreases both at low and high temperatures. The tropical and the temperate species show substantial differences in their temperature range for successful host location and also with respect to their performance at equal temperatures. Using laser Doppler vibrometry in *Pimpla turionellae* we can furthermore show a modification of the self-produced vibrational signals by the ambient temperature with respect to both frequency and intensity.

Hostile behavior to neighbors induced by encounter experience in the Japanese queenless ant, *Pristomyrmex pungens.*

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Ant workers cooperate among nestmates within colonies while they perform strong hostile behavior against non-nestmates. It is likely that neighbors are competitors for adjacent resources while strangers are transients from distant colonies. It might be adaptive for a species having the powerful competitors to discriminate neighbors from strangers and selectively attack the former. I hypothesized therefore that the workers can distinguish members of neighboring colonies from those of distant colonies based on some presumable cues in the ant *Pristomyrmex pungens.* To test this hypothesis, I created neighbors and strangers by an experimental method and compared the ant's hostile behavior toward the neighbors and strangers. The ants which had direct encounter experience with the neighbors could distinguish the neighbors from the strangers and showed more strong hostile behavior toward the former. The recognition of the neighbors was maintained for at least ten days after the most recent encounter. Nestmates without the encounter experience showed no hostile behavior toward the neighbors. This finding suggests that information about neighbors (probably colony odor) is not transmitted to nestmates within colonies. Aggressiveness toward neighbors therefore appears to be caused and maintained on the basis of individual memories from direct contact.

Insects associated with forest fires in northern California & Oregon.

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Fire has always been a component in the evolution of forest ecosystems affecting among other factors succession, nutrient cycling and species composition. Recent debate about fire suppression has highlighted a need for more information about how fire affects forest species. This study attempts to enumerate a diverse guild of insect species that appear to have coevolved with fire. Insects were hand collected at or shortly after approximately 40 forest fires that occurred in Northern California and Oregon in the latter half of the month of August between 1996 and 2003. Approximately 25 species in 8 families and 4 orders were collected. Factors affecting collection and distribution of insects at forest fires are discussed.

Infrared sensory systems in pyrophilous buprestid beetles.

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Jewel beetles of the genera *Melanophila* (distributed nearly all over the world except Australia) and *Merimna* (with the only species *atrata*, endemic to Australia) approach forest fires. The reason for this pyrophilous behaviour is that their xylophagous larvae only can develop in burnt wood. Both genera have infrared (IR) receptors to cope with their thermal environment. However, IR receptors are totally different in *Melanophila* beetles and in *Merimna atrata.* In *Merimna* 1 to 3 pairs of IR receptor can be found on the ventrolateral parts of the abdominal sternites. Receptors are innervated by a large multiterminal thermoreceptive neuron and function according to the bolometric principle. In *Melanophila* beetles, two pairs of metathoracic pit organs can be found directly behind the coxae of the middle legs. Each pit organs houses about 70 cuticular sensilla which function according to a photomechanic principle. Physiological and behavioural data suggest that *Melanophila* beetles may use their IR receptor for the detection of remote fires. On the other hand, *Merimna* may use its receptors in the near range to prevent to land on a hot surface. The fundamentally different structure and function strongly suggest that IR receptors have evolved independently in the two pyrophilous buprestid genera.

Fire recognition on the basis of insect olfactory systems.

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Insect antennae are morphologically and physiologically highly specialised organs for odour perception. Their sensitivity, selectivity, and response time cannot be matched by any technical method of trace analysis. Utilisation of these unique abilities of insects for trace analytical purposes demands a thorough adaptation of technical devices to the special needs of insect antennae. Yet approaches using electroantennogram (EAG) techniques are highly sophisticated, however, detailed examination of sensitivity and cross-reactivity of antennal responses to binary mixtures of odours are needed. By providing these data, practical applications of biosensors on the basis of intact insect antennae as a sensitive odour detecting and recognizing device become more feasible. Besides sex pheromones, insect antennae are able to detect a broad range of odours specific to their habitat. The Black jewel beetle (*Melanophila acuminata*) as well as the Australian pyrophilic beetle *Merimna atrata* shows a very high sensitivity to wood-fire specific volatiles. The results of first field tests of the biosensor will be reported. Practical application and technical development of the biosensor in fire early warning systems will be discussed.

Fire recognition based on new technologies.

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In Europe, huneds of human beings are killed and huneds of millions € damage to property are inflicted by fires in industrial, domestic or forestry areas every year. In many cases, losses of lives and property could have been prevented by efficient fire recognition and early warning systems. For this reason, development of fire recognition systems is a rapidly growing field integrating techniques as IR/vis- detection, inorganic and organic trace gas detection and aerosol analysis. Driven by results of biological research about fire recognition mechanisms used by animals, technical research and development was focussed to IR detection and organic trace gas detection i.e. "sensors for burnt smell". Especially tin oxide sensor elements showed favourable characteristics. Their high sensitivity and selectivity to hydroxylated and phenolic compounds in combination to low cross-reactivity with water and carbon monoxide mimics highly efficient biological fire detection systems. For sensor evaluation the HRGC/SOMMSA (High resolution gas chromatography/selective odorant measurement by multisensor array) approach has been established.

Pheromone trapping of *Halyomorpha halys* in the eastern USA.

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The brown marmorated stink bug, *Halyomorpha halys* (Heteroptera: Pentatomidae), is an introduced species in the USA; first being collected in eastern PA in 1996, and subsequently spreading to NJ and MD. We have observed this bug feeding on fruit, vegetables, soybeans, and many ornamental plants. Studies were initiated to evaluate the effectiveness of attracting and capturing *H. halys* in pheromone baited traps. Various purities of synthetic *Plautia stali* (2E,4E,6Z-1:COOMe) and *Thyanta pallidovirens* (2E,4Z,6Z-1:COOMe) (Heteroptera: Pentatomidae) aggregation pheromones were loaded onto rubber septa (2.5 mg/septum) along with a UV inhibitor and anti-oxidant. The first test was started on 12 September 2003. Individual treated septa were placed inside clear football-shaped traps that were hung in fruit trees near known *H. halys* infestations. *Halyomorpha halys* male and female adults and nymphs were captured in traps baited with *P. stali* or *T. pallidovirens* pheromone. A second test consisting of various purities (76, 88, and 94% pure) of *P. stali* aggregation pheromone and 2 types of traps was initiated on 6 October 2003. Football traps with the 76% purity pheromone captured more *H. halys* than traps with the 88 and 94% purity pheromone.

Contextual control of learning in insects.

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Insects are very adept at learning the association of many types of stimuli with biologically important events such as the occurrence of food, a mate, or a predator. There are several behavioral mechanisms associated with this type of learning, and they all in general allow animals to prepare for the occurrence of the event – e.g., flee, fight, or feed. Insects are equally adept at learning, through two or more means, to ignore stimuli that are not associated with important events. This prevents overuse of limited processing capacity associated with attention. With either type of learning, single stimuli are rarely perfectly reliable. The probability of an event (e) given the occurrence of a stimulus (s) - p[e|s] - might be above 0 but well below 1. But several stimuli together as a "context" might provide a more reliable predictor of an event. Associations among stimuli might underlie sensitive contextual control of a response. For example, in the context of S2 p[e|S1] might be much closer to 1 than in the context of S3. Furthermore, the meaning of S1 might be entirely different in S2 and S3. The presentation will review evidence for this type of contextual control of learning in insects.

Carrion mimicry in the dead horse arum.

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Some plants rely on deception for pollination. These plants typically mimic a favourable insect resource. Attracted insects are subsequently exploited as unrewarded pollinators. Here we describe the multisensory targeting mimicry of the dead horse arum, *Helicodiceros muscivorus*. The arum produces a characteristic odor, most similar to a carcass. Odor collections from the plant and from carrion contain the same active odor components, eliciting identical antennal responses in blowflies. Addition of these odor components to scentless flowers restores their attractiveness to the same level as plants with a natural scent. The arum is furthermore thermogenic, i.e. it produces heat. The arum is capable of producing over 10°C above ambient. Although, heat production is a fairly well known phenomenon, few studies have provided direct experimental evidence as to what function heat plays. We have shown that the heat in the arum primarily serves as a behavioral altering stimulus, enhancing threefold the likelihood that the flies will enter the trap chamber of the plant. Accompanying the olfactory and thermal cues are also visual and tactile cues, reinforcing the overall carrion appearance. The dead horse arum serve as a striking example of how a plant during evolution can acquire the ability to mimic multiple aspects of complex objects.

Pheromone & visual factors contribute to catch of *Synathedon tipuliformis.*

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Currant clearwing *Synanthedon tipuliformis* (Sessiidae) has been a pioneering and successful target of mating disruption in New Zealand, widely adopted since c. 1990. However, control failures led us to examine pheromone loading and coloured traps. Male attraction to yellow delta traps was equivalent to green delta traps but was greater than to red, black, blue or white traps. Fully yellow delta traps were more attractive than black traps with yellow stripes designed to mimic the insect, and it is suggested that yellow provides a supernormal stimulus (female abdomens have two yellow stripes). However, there was no catch in the absence of the pheromone lure, showing that colour alone was not a sufficient stimulus. Catch increased as a function of pheromone loading and trap colour, but the effect appeared to be additive. However, when plastic funnel traps were used, catch in yellow traps was highly significantly greater than in green plastic traps, indicating greater discrimination between these colours. Curiously, a composite coloured lid-funnel-base of green-yellow-white funnel traps was more attractive than fully yellow-yellow-yellow coloured funnel traps or composite green-yellow-green. These results suggest that the insect has a high degree of visual discrimination between colour shades and surface area. While significant improvements (c. 16-fold) to catch have now been achieved, with an additive combination of trap colour and pheromone loading, more work is needed to decipher the visual discrimination in this species.

Putative identification of the contact sex pheromone of the burnt pine longhorn, *Arhopalus tristis* (Coleoptera: Cerambycidae).

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The longhorn *Arhopalus tristis* (F) is a pest of pines, particularly those damaged by fire, and is a major export quarantine issue in New Zealand. In our project to develop an attractant to help in monitoring and management of this species, we have investigated the contact sex pheromone. Male burnt pine longhorn initiates copulatory responses to females only after sensing them with their antennae, providing evidence for contact sex pheromone in this species. When applied to a gelatine capsule, hexane wash of female's elytra elicited male copulatory attempts similar to the copulatory response elicited in males by live females. GC-MS analysis of male and female elytra extracts indicated that all of the compounds present in both extracts were saturated and branched hydrocarbons from 25Hy to 33Hy. The GC-MS analysis of the hexane extracts of male and female elytra revealed that the profile of compounds in both extracts was qualitatively similar, but different in the ratio of two peaks. These two peaks were identified as branched 28Hy hydrocarbons and a straight chain 29Hy. Further work is underway to test the activity of these compounds.

Genetic variation in learning ability may mask population-level effects of experience.

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Variation in learning abilities has long been suspected to have a genetic basis and recently experimental evidence in support of this hypothesis is accumulating. In spite of these developments, in many studies of cognitive abilities such a variation is not controlled for, because populations of mixed or unknown genetic background are tested. We postulate that such an approach may confound the occurrence and/or magnitude of learning effects. We support this postulate with our results on learning in the predatory mite *Phytoseiulus persimilis*. *Phytoseiulus persimilis* responds to volatile chemicals released by plants following infestation by its prey, the two-spotted spider mite. The blend of induced volatiles is herbivore-specific and as such it serves as a signal of prey presence. It has therefore been predicted that learning of volatiles would be highly beneficial for this mite. We show that a collection of *P. persimilis* from natural populations on Sicily exhibits no significant change in responses to the volatiles following the experience. The same tests, however performed on iso-female lines from those populations show variation in learning ability among the lines. These as well as other results on variation in learning abilities raise the question what mechanism maintains such a variation.

Specificity of herbivore-induced volatiles in information transfer in multitrophic interactions.

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When infested by diamondback moth (DBM) (*Plutella xylostella*) larvae, cruciferous plants emit volatiles that attract a specialist parasitic wasp of DBM larvae, *Cotesia plutellae*. The wasps are not attracted to volatiles emitted by a crucifer plant infested by nonhosts [cabbage butterfly (CB) (*Pieris rapae*) larvae]. We identified specific volatile compounds emitted by cabbage plants with minor DBM larvae infestations, but not emitted by intact cabbage plants. When we compared the attractive properties of each chemical on an intact cabbage plant (treated plant) with that of the solvent alone (control plant), the wasps were equally distributed between the two plants. However, when a mixture of all the chemicals was used instead of each chemical alone, the wasps showed a significant preference towards the treated plants. These data suggest that a specific blend is important in the interaction between cabbage plants, which are infested by DBM larvae, and *C. plutella*. By comparing this tritrophic system with one consisting of cabbage plants, CB larvae, and their parasitoid *Cotesia glomerata*, the specificity of herbivore-induced volatiles in information transfer is investigated.

Cone thermogenesis & volatiles associated with specific pollinators of *Macrozamia* cycads.

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Macrozamia cycads, endemic to Australia, are associated with an unusual and diverse array of insect pollinators compared with other cycads. Some *Macrozamia* species are pollinated by *Tranes* weevils only, others only by thrips (*Cycadothrips)*, and some by both insect types. We show that these cycads' cones have chemical and metabolic traits that are associated strongly with their specific pollinator type. Species pollinated by *Tranes*- only have cones that heat and release volatiles during the early evening, whereas *Cycadothrips*-only cycads have cones that heat and release volatiles during the day. Cones of *Tranes*-pollinated species release volatiles comprising a few monoterpenes, primarily linalool and much lower levels of beta-myrcene, whereas "thrips-only" cycads lack linalool and release numerous monoterpenes, dominated by beta-myrcene emitted at very high rates. Insect movement between male and female cones occurs during the cone heating and volatile emission phase of their specific host. Preliminary evidence suggests that insects are attracted to their host's cone volatiles and they respond neurologically to specific components within the volatile mix.

Cuticular hydrocarbon patterns of termitophiles & their host.

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Termitophile can be defined as any animal other than a termite that is normally found in a termite nest. Termitophiles supposed to use "appeasement chemicals" to integrate into the social structure of termite colonies. Since cuticular hydrocarbons are caste- and species-specific, these mixtures may important to termite, possibly used as species recognition cues. Termitophiles were collected from termite nest or around termite galleries in Taiwan. Using gas chromatographic analysis to compare the cuticular hydrocarbons of some termitophiles and their hosts, the chromatograms show some cuticular hydrocarbon components of termites might be similar to those of their termitophiles. Termitophiles might utilize chemical mimicry to integrate into termite society. The similarity of cuticular hydrocarbon component between termitophiles and termite host might indicate the integration degree of termitophiles.

Perspectives for time allocation studies in the future.

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The allocation of foraging time in insects has been a fruitful area blending empirical and theoretical research. About 100 papers with insects or mites as study subjects have appeared since 1990, the same number as for all other organisms (birds, fishes, mammals and gastropods). A survey of this literature and recent developments point to the following promising areas of research.
(A) Recent models on group foraging have not yet seen empirical tests
(B) Individual variation in behaviour has been amply documented. Three explanations have been forwarded to explain this variation: (1) information acquisition and learning, (2) differences in internal state and (3) genetic variation within and between populations. However, the relative importance of these sources of variation has not been investigated.
(C) The recent finding that symbionts, like bacteria and viruses, can change parasitoid behaviour raises the question, of which behaviours might be extended phenotypes of these symbionts, and to what extent conflicts between symbionts and their hosts occur.
(D) Host foraging, the most studied process in patch allocation is one facet of different trade-offs, such as searching for food and searching for hosts. Little is know on how these trade-off affect parasitoid foraging time and how the animals integrate the information of the presence of different resources.
(E) Up to now the study field of patch residence time is restricted to behavioural macroscopic, processes. Insect physiologist should be attracted to the field to unravel processes as the integration of different signals, how the time elapsed is integrated to patch leaving mechanisms, etc.

A maternal chemical cue affecting the behaviour of a parasitoid of aphids on emergence.

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A fully-defined artificial diet containing no secondary plant compounds produces aphid mummies which give emerging parasitoids no opportunity to learn plant volatiles of the plant on which they were reared. To improve parasitisation of the aphid *Myzus persicae* (Sulzer) by the generalist parasitoid *Aphidius colemani* (Viereck) on the diet, we allowed parasitisation on a host plant leaf for 48 h, after which the aphids were returned to the diet. We now needed to test that this procedure did not provide plant volatile cues to the emerging parasitoids. Unfortunately it did, but only when the plant from which the parent parasitoids had been reared was the same as that on which they had parasitised the aphids. Thus, if parasitisation has occurred on Brussels sprouts, the emerging daughters only preferred the odour of sprouts to cabbage if their parents had been reared on sprouts; if reared on cabbage, there was no olfactory preference for either brassica. The parasitoids clearly leave a maternal cue for their offspring when they oviposit, and we discuss what this might be, where it might be located in the parasitised aphid, how it affects the emerging offspring and why it is left.

Sexual behavior in *Nezara viridula* (L) (Heteroptera: Pentatomidae).

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The southern green stink bug *Nezara viridula* (L.) (Heteroptera: Pentatomidae) has a complex sexual behavior. As a part of a specific mate recognition system, during courtship male and female exchange chemical and substrate-borne vibrational signals. Sexually mature males release a volatile sex pheromone that is regarded as a long-range attractant and serves to bring bugs to the same plant. When on a plant, both male and female emit several species and sex specific vibrational signals called songs. It seems that volatile chemical signal triggers the emission of the female calling song. Male responds to female calls with his songs and he locates the singing female on a plant using her vibrational signals as a directional cue. Male songs keep female singing for a longer time with a regular repetition rate while the female calling song modulates the emission of the male sex pheromone. Such feed-back modulatory mechanisms probably help to improve the synchronization of sexual activity and could also provide the sensory cues for sexual selection.

Mating behaviour of *Zorion guttigerum* (Westwood) (Coleoptera: Cerambycidae).

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Zorion guttigerum *(Westwood) is a diurnal cerambycine beetle endemic to New Zealand. Pair-bonding, mating, and adult feeding occur on flowers while larvae feed on dead tree twigs. Adults live for > 3wks and the male and female arrive at flowers independently, suggesting that long-range sex pheromones may not operate in mate location. Mean pair-bonding period is 22.7± 24.2 (mean ± SD)(ranged from 2.3 to 120.5) min during which time 1.8 ± 1.5 (ranged from 1 to 6) matings occur. Mating lasts an average of 4.7± 2.5 (ranged from 1.2 to 11.7) min. Overall operational sex ratio is male-biased (»1♀:1.5♂) but it becomes very highly male-biased (»1♀:9♂) when pair-bonding and feeding activities decrease to the minimum between 1400 and 1600 h, suggesting that females leave flowers for oviposition. It is probably true that for cerambycid species whose females oviposit alone and mating and oviposition occur on different plants or different parts of plants, the operational sex ratio varies significantly over time on the mating/feeding sites. Duration of pair-bondings also varies over time. Shorter pair-bondings in the morning may suggest a strong sexual selection process. After ca. 2-h selection, both sexes tend to engage in much longer pair-bonding and mate more times before females leave the mating and feeding sites for oviposition in the early afternoon.*

Towards the identification of contact pheromones of the fig longicorn, Agrianome spinicollis *(Macleay).*

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Agrianome spinicollis is a cerambycid endemic to the eastern states of Australia and is a serious pest of pecan nut trees. Damage results from larvae feeding within the limbs and trunks of the trees, which ultimately results in the trees' collapse. Observations suggest that contact pheromones on the elytra of female beetles form an important component of the mate recognition system employed by male beetles. Experiments were conducted in which the cuticular hydrocarbons were removed from both male and female beetles. Such solvent-rinsing of females resulted in the loss of attractiveness towards males. The subsequent re-application of the hexane rinse, at concentrations as low as 0.1 female equivalents, was followed by mating attempts by males. Gas chromatographic analysis of the cuticular rinses revealed significant quantitative sex differences in shared components, and possibly qualitative differences. The male extract is dominated by mono-unsaturated hydrocarbons having 25, 27 and 31 carbons. The female extract, in contrast, is dominated by a single alkene, with 26 carbon atoms. As a result of the short range of efficacy of the compounds, these findings are unlikely to have any significant management implications.

Influence of lek size on female visitation in Queensland fruit fly (*Bactrocera tryoni* Froggatt) (Diptera: Tephritidae).

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The lek mating system of Queensland fruit fly, *Bactrocera tryoni* (Froggatt) (Diptera: Tephritidae), may reduce the ability of this species to colonise new areas. The importance of lek size for female visitation at lek sites and initiation of male pheromone-calling was investigated in large cages using artificial leks. Female visitation at artificial leks was monitored to determine if females visited leks due to passive attraction or showed a preference for larger leks. Female *B. tryoni* display a preference for larger leks rather than being passively attracted to a greater perceived stimulus from large leks. Per capita success of males in attracting females was higher at larger leks than where there was a single male. Calling propensity of male *B. tryoni* was increased by the presence of conspecific males. Overall, female visitation at leks was only weakly associated with male calling, suggesting that lek size and the number of pheromone-calling males is not the only factor important in locating mates in this species.

Integration of multi-sensory information in orientation & navigation behavior.

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The ultimate goal of neuroethology is to describe the neural mechanisms that produce adaptive behaviors in freely moving animals. My lab has been using a virtual reality flight system that closely emulates realistic stimuli that occur during flight, and which produce robust flight behaviors in a tethered moth (*Manduca sexta*) that are similar to those seen in real flight. Concurrently we are able to utilize extracellular multi-unit electrodes to record activity in the region of the ventral nerve cord between the brain and the thoracic ganglia. This system allows us to investigate mechanisms by which *Manduca* responds to visual and olfactory stimuli that it may encounter during actual pheromone-guided flight, and how it might activate and control flight manoeuvres. Initial results reveal ensembles of neurons that show unique activation patterns associated with visual pattern movement associated with flight, and that these patterns are modulated during exposure to pheromone. Similarly, other unique ensembles of neurons are activated differentially when the preparation is in tethered flight or not flying. These experiments are aimed at understanding how multiple streams of information are integrated by the nervous system and how this is meshed with ongoing behavior. Supported by: ONR-MURI No. N00014-01-1-0676.

Pheromone-modulated orientation mechanisms in walking male cockroaches.

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Perhaps the most remarkable olfactory task achieved by insects is that of locating distant resources using airborne chemical cues. It is currently thought that two primary mechanisms underlie flying insects' ability to perform this behavior: 1) an odor-modulated, visually-mediated orientation to the wind direction and, 2) an odor-activated internal program of turns. The blending of these two mechanisms is thought to generate the upwind zigzagging paths that we observe from insects tracking odors in flight. It is not yet clear whether vision contributes significantly to the odor tracking behavior of walking insects. Recent results suggest that visual information may be very important, but not used in the same manner as flying odor tracking insects. Furthermore, the structure of the walking paths of pheromone tracking cockroaches look remarkably like those observed from flying insects, and thus may be governed by similar mechanisms. Our new results suggest that comparing the odor tracking behavior of walking and flying insects in similar environments may reveal important similarities and differences in how olfactory and other sensory information is used. Supported by: ONR-MURI No. N00014-01-1-0676.

Odor concentration & odor invariance in the olfactory system of the honeybee.

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Sensory systems have evolved to recognize stimuli over a range of stimulus intensities. A stimulus may be more intense, but it will still retain other qualities that make it classifiable along another dimension. When the qualitative representation of a stimulus remains the same over a specific range of intensity, the stimulus is defined as *invariant* over that range. In olfaction, it is often noted that increasing the intensity of an odorant affects its quality, such that odorants do not remain invariant as a function of their concentration in air. I will discuss how our experiments with honeybees reflect the extent that odorants remain invariant and the implications of failure of invariance for odor coding.

Local resource competition in the solitary parasitoid *Haplogonatopus atratus* (Hymenoptera: Dryinidae).

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This is the first study showing local resource competition (LRC) in parasitoids. Females of the parasitoid *Haplogonatopus atratus* are apterous, while males are alate. Individuals of the host, *Laodelphax striatellus* (Hemiptera Delphacidae), are distributed in clumps in paddy fields. These facts suggest the existence of LRC among female offspring of mother parasitoids, and that female parasitoids will produce a lower proportion of male offspring in patches with higher host density. Our laboratory experiments confirmed these predictions, as follows. A female parasitoid was placed in a glass pot with rice seedlings on which different numbers of planthoppers (10, 20, 40, or 80) were feeding. The female was allowed to oviposit freely until she left the seedlings. Sex ratios (male proportions) in emerging adults decreased with increasing host density. Moreover, the average number of eggs laid among parasitized hosts decreased as the host density increased, but hosts with different numbers of eggs laid in the same patch produced similar sex ratios. Host-quality models suggesting that male eggs are more likely to be laid in hosts of low quality (i.e., the parasitized hosts in this case) do not explain the above patterns in the sex ratio.

Identification of semiochemicals for the Asian longhorned beetle, *Anoplophora glabripennis.*

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A series of long-chain hydrocarbons comprise the cuticular waxes of both sexes of ALB adults. Although for the most part the gas chromatographic profiles are similar for the two sexes, five monounsaturated compounds were consistently more abundant in samples from females than in those from males. These compounds were identified as (*Z*)-9-tricosene, (*Z*)-9-pentacosene, (*Z*)-7-pentacosene, (*Z*)-9-heptacosene, and (*Z*)-7-heptacosene in the approximate ratio of 1:2:2:8:1, respectively. Antennal contact to a micro-centrifuge tube coated with synthetic mixture of the five compounds stimulated copulatory behavior in males. This is the first time that monounsaturated hydrocarbons, (Z)-9-tricosene, (Z)-9-pentacosene, (Z)-7-pentacosene, (Z)-9-heptacosene, and (Z)-7-heptacosene, have been implicated as contact sex recognition pheromone components in the family Cerambycidae. Two male-specific beetle volatiles were found that elicited strong gas chromatographic-electroantennographic detection (GC-EAD) responses from both sexes of adult Asian longhorned beetle (ALB), *Anoplophora glabripennis*. The secretion consisted of a ~1:1 blend of functionalized dialkyl ethers, 4-(*n*-heptyloxy)butanal and 4-(*n*-heptyloxy)butan-1-ol. These compounds are chemically unusual natural products that are previously unknown from insects. Laboratory olfactometer studies showed that a blend of 10 µg of each synthetic compound on a filter paper strip was significantly attractive to ALB adults.

Identification of sex pheromones of *Holcocerus hippophaecolus* (Lepidoptera: Cossidae) & biological activities.

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Holcocerus hippophaecolus Hua (Lepidoptera: Cossidae) is a destructive forest pest widely distributed throughout northern and western region of China. Its primary host-plant is sandthorn (*Hippophae rhamnoides* L.), which is a most widely planted shrub used for preventing from soil erosion and desert control, along the area of loess plateau in China. The fruit of sandthorn could be used for juice and Chinese medicine. Its plantation reached 180 million ha. About half million ha of plantations suffered severely attacking by the pest insect. Seven components were identified from female sex pheromone gland. These components were (*E*)-3-tetradecenyl acetate (E3-14:Ac), (*Z*)-3-tetradecenyl acetate (Z3-14:Ac), (*Z*)-7-tetradecenyl acetate (Z7-14:Ac) and related alcohols: E3-14:OH, Z3-14:OH, Z7-14:OH and (*Z*)-9-tetradecenyl acetate (Z9-14:Ac). Two major ones are as Z7-14:Ac and E3-14:Ac. EAG showed that Z7-14:Ac produced the strongest response and followed by E3-14:Ac in these seven compounds. The field trials showed that the combination of Z7-14:Ac and E3-14:Ac in a ratio of 1:1 could significantly catch much more males than others.

First identification of the sex pheromone of an Australian Lymantriidae, *Teia anartoides:* theromo-labile diverse pheromone blend.

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Active site of AnyParP, a peptide inducing embryonic diapause of *Bomby mori*

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AnyParP (*Antheraea yamamai* paralytic peptide) is a 23-residues peptide with one disulfide bridge. Recent studies showed that synthetic AnyParP not only paralyzed the larvae of both *A. yamamai* and *Bombyx mori*, but also induced embryonic diapause of *B. mori*. To identify the residues important for diapause egg-inducing activity in polyvoltine race (N4), *B. mori*, a series of synthetic mutants of AnyParP were used in bioassay system for comparing with the wild-type AnyParP. Ala replacements of Cys^7 and Cys^{19} (C7.19A), Glu^1-Asn^2 (AA-AnyParP), as well as Phe^3 (F3A) eliminated all activity. The present results indicate that the disulfide bond between Cys^7 and Cys^{19} is essential for maintaining the three-dimensional structure of AnyParP and that the E^1, N^2 and F^3 are very critical for the diapause egg-inducing activity of AnyParP. Our results indicate that these residues in both the core and unstructured domains of AnyParP are required for diapause egg-inducing activity.

Comparision of development of bean bug, *Riptortus clavatus* Thunberg (Hemiptera: Alydidae) and yellow-brown stink bug, *Halyomorpha halys* Stal (Hemiptera: Pentatomidae) at temperature conditions

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Development of Bean bug (BB), *Riptortus clavatus* Thunberg and Yellow-brown Stink Bug (YBSB), *Halyomorpha halys* Stal, which are considered as major sucking insect pests on soybean and fruits such as sweet persimmon and apple in Korea, were observed at four temperatures (20, 24, 28 and 32°C). Egg duration was significantly longer in BB (7.0 to 16.7 days) than that in YBSB (3.9 to 12.5 days). At 20, 24 and 28°C, nymphal durations of BB and YBSB were in the range of 23.0 to 37.8 days and 29.9 to 65.0 days, respectively. At 32°C, however, nymphal duration of BB was 18.1 days while all nymphs of YBSB were died within 2nd instar. Pre-oviposition of BB was greatly shorter with 5.4 to 10.8 days than that of YBSB with 9.1 to 43.1 days. Adult female longevity of BB was the longest with 49.0 days at 28°C and the shortest with 12.8 days at 20°C while YBSB was the longest with 75.3 days at 20°C and 28.2 days 28°C, adversely. Accordingly, one generation period of BB and YBSB was 67.3, 79.6, 80.7 and 44.1 days at 20, 24, 28 and 32°C, and was 152.8, 102.4 and 62.6 days at 20, 24 and 28°C, respectively. The threshold temperature was 9.6°C in egg and 9.8°C in nymph of BB while was 13.8°C in egg and 13.3°C in nymph of YBSB.

Summary diapause of *Chrysocharis pubicornis*, a pupal parasitoid of the garden pea leafminer, *Chromatomyia horticola*.

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The occurrence of summer diapause in the last larval instar of *Chrysocharis pubicornis*, an Eulophid pupal parasitoid of Agromyzid leafminers, was studied in the laboratory. The effects of three constant temperatures (15, 20 and 25°C) and three photoperiods (12, 14 and 16 h) on diapause induction and termination were evaluated in different combinations. Diapause induction was found to be under control of photoperiod and temperature. Larval development showed a short-day type response; the lowest diapause induction rate (0.67%) occurred at 15**°C** and 12L:12D, and gradually increased with photoperiod and temperature, reaching the highest value (74.9%) at 25**°C** and 16L:8D. The critical photoperiod was between 14 and 16h at 25°C. For diapause termination, regardless of photoperiod, the lowest temperature (15°C) caused diapause termination in 90% of individuals, whereas the termination rate at the highest temperature (25°C) was practically 0%; it was only at 20°C when photoperiod exerted effect, with diapause termination nearing 90% at the short photoperiod (12 h) but only 16% at the longest photoperiod (16 h). The adaptation of *C. pubicornis* to the population trend of *C. horticola*, its main host in Japan, as well as the possible implications of summer diapause in a local Agromyzid-leafminer biological control program in greenhouses are discussed.

Can the ligand binding domain of *Xenos peckii* USP/RXR be used as an indicator of strepsipteran phylogenetic affinities?

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The receptor for the insect molting hormone, ecdysone, is a heterodimer consisting of the Ecdysone Receptor and Ultraspiracle (USP) proteins. The DNA-binding domains of arthropod USPs and their vertebrate homologs, the retinoid X receptors (RXRs) are highly conserved. The ligand binding domain sequences, however, divide into two distinct groups. One group consists of sequences from members of the holometabolous Lepidoptera and Diptera, while the other available arthropod sequences group with vertebrate RXRs. Because the separation between these two groups is so distinct, we wondered whether USP/RXR structure could be used as a tool for clarifying the contentious phylogenetic position of the Order Strepsiptera, which has been placed as sister to either Diptera or Coleoptera. We have cloned and sequenced the USP/RXRs from the Strepsipteran *Xenos peckii* and its host *Polistes fuscatus* and bioinformatic analyses are currently in progress. The results will be reported

Interaction of juvenogens & insect oostatic peptides with developing ovaries of *Neobellieria bullata* (Diptera)

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Juvenogens, fatty acid esters of selected insect juvenile hormone bioanalogues, and insect oostatic peptides (H-YDPAP-OH and H-YDPA-OH) were selected from the screening programme on a model object *Neobellieria bullata* (Diptera) as the most promising ones. They were applied by injections (peptides) or by topical application (juvenogens). At given intervals the flies were dissected and their ovaries were examined for morphological changes and hatchability of eggs in the uterus, respectively. Under described regime of applications the development of the first egg was not morphologically affected. Hatchability of eggs in uterus slightly decreased, ranging from 60 to 80 percent compared to controls. The second gonotrophic cycle was characterized by remarkable changes in the follicular epithelium of about 60% of oocytes. Monolayered cylinical follicular epithelium with nuclei in the centre is altered the following way: first the nucleoli became prominent and subsequently multiplied, nuclei show slight difference in their size. Some bizarre nuclei with great number of nucleoli start to appear; nucleoli often merge together. The nucleocytoplasmatic ratio grows for the benefit of nucleus. Groups of giant cells appear. The epithelium proliferates towards the inner part of egg chamber. In some sections the follicular epithelium forms pseudopapillar projections, stratification is severely disturbed and extensive invagination to the centre of the egg chamber appears.

Life cycle of *Hydroscapha natans* & the development of gonads

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Hydroscapha natans belongs to the myxophagan beetles (Coleoptera, Myxophaga), which is one of the "ancient" suborders of beetles. *H. natans* live in shallow water habitats and feed on algae. I have bread this species successfully in the laboratory for several years. After copulation females lay eggs over a period of one to two month. Two eggs can be layed daily. Embryogenesis: 5d, the primary larva (L1): 3d, L2: 3d, L3: 4d, L4: 11d, pupal phase: 2,5d. Live span of adults is about half a year. The gonads are formed during larval stages. In males the testes are composed of three testioles each. The polarized testioles house anteriorly stem cells, cystoblasts and mitotically dividing cysts. The zone of meiotic divisions and finally the zone of spermiohistogenesis follow. In females, three telotrophic meroistic ovarioles build each ovary. The morphology is similar to the telotrophic ovaries found as yet only in Neuropteroidea (Megaloptera, Sialidae and Rahphidioptera). During larval phases, ovarioles develop small germ line cysts in their anterior compartments. Later, membranes of central cyst cells dissolve and a membrane free central syncytium remains surrounded by a monolayer of germ cells, the tapetum cells. Only the posterior tapetum cells can develop subsequently into oocytes.

Cadmium effects on vitellogenesis in two strains of *Oncopeltus fasciatus* (Heteroptera: Lygaeidae)

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Cadmium may act as an endocrine disruptor in fishes, and also can inhibit vitellogenin (Vg) synthesis in the large milkweed bug *O. fasciatus*. Vitellogenin (VgA and VgB) levels in hemolymph of 3 and 5-day old insect females exposed to 50 mg Cd/litre in inking water were determined by SDS-PAGE. Two different strains were studied: OS, a laboratory-reared strain, and OC, a strain that had been subjected to Cd selection for more than 40 generations, and supported 50 mg Cd/litre at the moment of the experiment. Vitellogenin levels in OC females were significantly higher than those in OS females (88% for VgA and 44% for VgB at day 3, 16% for VgA and 22% for VgB at day 5). When OC and OS females were exposed to 50 mg Cd/litre, Vg concentrations were decreased in both strains. However, Vg levels in OC exposed females were still similar to those of control OS females. Likewise, OC females chronically exposed to 50 mg Cd/litre from eclosion to adulthood had reduced levels of hemolymph Vg when compared to control OS insects, but Vg concentrations were higher that those of OS insects exposed to 50 mg Cd/litre for 2-5 days. OC females also accumulated 20% more Cd than OS females. These results suggest that enhanced Vg levels may play a role in Cd resistance in the OC strain.

The expression of circadian rhythm in electroantennogram (EAG) of the German cockroach, *Blattella germanica* (L.)

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Male German cockroaches (*Blattella germanica*) express circadian rhythmicity in locomotion and its peak activity occurs at night. We found circadian rhythm of neuron activity expressed in olfactory response by measuring electroantennograms (EAGs). By using ethyl acetate as the stimulus, the amplitude of EAG response kept fluctuating in subjective day and night under constant darkness. Although female German cockroaches do not express locomotion circadian rhythm, the EAG responses of female expressed robustly. The peak of response was at the beginning of subjective night. Dilution-responses curves of EAG indicated that the amplitude of 10-fold different dilutions showed only 3 times of change in sensitivity. The results indicated that both sexes expressed EAG circadian rhythm and masking effect on female's locomotion did not hinder the EAG response.

Clock controlled proteins from the vas deferens of *Spodoptera littoralis*

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The circadian oscillator located in the male reproductive tract of *Spodoptera littoralis* controls sperm release from the testis into the upper vas deferens (UVD). The daily cycle of sperm release is precisely regulated; sperm leave the testis at dusk, and remain in the UVD for several hours. The retention of sperm in the UVD is associated with secretion of glycosylated proteins from the UVD epithelium. The goal of this study was characterization of these rhythmically secreted glycoproteins. Protein extracts from the UVD wall, luminal fluid and sperm were analyzed using 2D-electrophoresis. Two acidic 105kD and 40kD glycoproteins were detected in the UVD lumen prior to sperm release while two other proteins of 50kD and 75kD were detected during sperm release. Differences were also observed in the glycoprotein composition between pre-release sperm from the testis and sperm in the UVD lumen. Taken together these data suggest that rhythmic sperm release is associated with glycosylation of existing sperm proteins and attachment of new glycoproteins to the spermatozoa in the UVD. Using lectins to characterize protein glycosylation in UVD tissues and sperm, it was shown that the glycan chains of these proteins are chiefly carbohydrates with terminal mannose and glucose residues. Supported: KBN2P04C04426.

Egg maturation & bacteriocyte inclusion in immature & adult *Bemisia gigantea* Martin (Hemiptera: Aleyrodidae)

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In this study we investigated patterns of egg production and inclusion of bacterial endosymbionts in the whitefly *Bemisia gigantea*, a species native to lowland tropical rainforests of North Queensland, Australia. Development of the female reproduction system of *B. gigantea* becomes evident in fourth instar nymphs, with extensive accumulation of mature (chorionated) eggs occurring 1-2 days prior to adult eclosion. Evidence from light and electron microscopy indicates that bacteriocytes (specialised cells containing bacterial endosymbionts) appear scattered through the abdominal region in close association with the developing eggs. In fourth instar nymphs, bacteriocytes enter developing eggs basally through the pedicel, and after oviposition move distally from the pedicel as the embryo matures. In addition, developing ova without bacteriocytes were evident in 3-day-old females, indicating that oogenesis and bacteriocyte inclusion is an on going process.

Adenosine deaminases

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We present a comparison of insects and human adenosine deaminases, their phylogenetic relationship, expression patterns and possible roles in organisms based on lost of function mutations. Adenosine deaminases are involved in the enzymatic conversion of nucleosides adenosine and 2`-deoxyadenosine into inosine and 2`-deoxyinosine, respectively. Recent discoveries and completion of genome sequencing of several model organisms revealed that two subfamilies of adenosine deaminases with common enzymatic domain but different N- terminal regions could be distinguished among metazoa. The first group contains „bona-fide" adenosine deaminases, while the second group is comprised of ADGF-like adenosine deaminases. Most species examined, including human and *D. melanogaster* contain genes belonging to both subfamilies. Vertebrate and invertebrate adenosine deaminases are subject to complex developmental and tissue-specific regulation. Despite the fact that mammals and insects prefer different subfamily members as "principal" adenosine deaminases, an interesting parallel exists in both groups in respect of the requirement of highest adenosine deaminase activity – in guts and lymphoid organs/blood cells.

Protein expression in the reproductive organs of male German cockroach (*Blattella germanica*)

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Male accessory reproductive glands (ARGs) contain factors that are transferred with sperm during mating to the female. One such factor (sex peptide of Drosophila) inhibits female's receptivity to additional mating. Here, we analyzed ARGs protein content and pattern for males of the German cockroach (*Blattella germanica*). Lower protein mass were found in young males (<3day-day-old) when compared to older ones (3~10-day-old) by SDS-PAGE. This study shows the expression of genes in ARG was different with age, but the protein content of testis remained the same after adult emergence. In addition, western blot analysis and whole-mount immunohistochemistry were used to investigate sex peptide-like immunoreactivity in the reproductive organs of the male. Unexpectedly, the sex peptide-like substance was only found in the testis of *B. germanica*. The significance of this finding is currently under investigation.

Local adaptation & diapause traits of two noctuid moths, *Helicoverpa armigera* **&** *H. assulta* **(Lepidoptera: Noctuidae)**

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In recent years, two noctuid pests are becoming more and more hazardous in agroecosystems in Japan. One is the cotton bollworm, *Helicoverpa armigera*, which is rather described as a newcomer species from the south, while the other, the oriental tobacco budworm, *H. assulta*, has been observed for decades in Japan. When focused on their local adaptations, the ability to overwinter is absolutely vital, although it still remains uncertain how the ability contributes in the field or how it differs among species. Preceding researches reported no geographical cline in photoperiodical reaction of local populations of *H. armigera* in Japan. Even less attention has been paid for *H. assulta*. On the way of our research, it turned out that diapause reaction to a constant photoperiod was not a reliable standard to clarify interspecific or interpopulation variation in diapause tendencies of these noctuid moths. It should be desirable that standard methods are more reflective of natural conditions such as decreasing temperature or shortening photoperiod through the development period. Reconsideration of diapause traits of these species revealed detailed differences that stem from the specific climate and their distinctive biology, which will lead to deeper understanding of their local adaptation.

Insect hormones, theirs analogues & antagonists in development & reproduction of *Spodoptera littoralis* (Boisd)

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Effects of methoprene, 20-hydroxyecdysone, metyrapone RH-2485 and RH-5992 on the development and maturation of male germinal cells in *Spodoptera littoralis* were studied. Spermatogenesis begins early in the postembryonic development. In the first to third larval instars formation of spermatogonia occurs. Meiotic division starts in 4th instar and in the last larval instar all developmental stages of spermatozoa can be found. The first bundle of mature spermatozoa occurs in prepupae. In 5 day-old-pupae more than 90 % of the spermatozoa are mature. Application of juvenoids or ecdysterone caused some disorders in the development spermatozoa. After application of different concentration of methoprene on the different developmental stages reduction of the reproductive ability of males was observed. This reduction is caused by numerous disorders in development of spermatozoa. Numerous pycnosis appear in spermatogonia, inhibition of spermatocytes and their gradual disintegration and disorders of spermateliosis take place. On the other hand application of 20-hydroxyecdysone pathological changes were not found and the spermateliosis was earlier with comparison with control animals. After application of metyrapone or RH compounds the pathological changes were not found and experimental animals were fully fertile. All tested compounds caused dose dependent changes in the development – prolongation of larval intermoult periods, formation of different larval pupal intermediates.

Heat-shock-responsive genes are not involved in the expression of diapause in two dipteran species, *Drosophila triauraria* & *Lucilia sericata*

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Although the molecular regulation of diapause remains largely unknown, there is an accumulation of data suggesting the involvement of heat-shock proteins (Hsps) in the expression of diapause or dormancy. However, we previously reported that Drosophila *triauraria* Bock & Wheeler (osophilidae) does not express *Hsp70* transcripts at normal temperatures, regardless of the diapause state. Here we investigated the transcriptional regulation of heat-shock-responsive genes, *Hsp23*, *Hsp26*, *Hsp83* and *Hsrw* in diapause adults of *D. triauraria* and *Hsp23*, *Hsp70* and *Hsp83* in diapause larvae of *Lucilia sericata* (Meigen) (Calliphoridae). The present study revealed that these genes are not regulated as a function of diapause, suggesting that they are not involved in the expression of diapause in both species. It is noteworthy that in *L. sericata Hsp83* expression was clearly up-regulated when diapause was terminated by temperature- and/or long-day-exposures. The results suggest that expression of *Hsp83* is a part of the process promoting normal development.

Electron-tomography of the adult retinal cells in *Drosophila melanogaster.*

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Studies about the structure of *Drosophila melanogaster*'s photoreceptor cell using electron microscopy were carried in detail by Waddington (1962), Farquhar and Palade (1963), and Perry (1968). But, these results can have limitations in functional research because of two-dimensional structure. High voltage electron microscopy (HVEM) has been a natural outgrowth of the desire to obtain 3-dimensional information due to problems related to interpretations of 3-dimensional images from 2-dimensional electron microscophs are numerous. The fine-structure preservation of cells or tissues can be achieved by using fast freezing followed by freeze-substitution techniques. It is estimated that samples prepared by high pressure freezing are fixed in 20-50 mess, as compared to the seconds and/or minutes by chemical fixations. In this study, the adult retina of *Drosophila melanogaster* was investigated employing HVEM, fixation by high pressure freezing followed freeze-substitution, thick serial sections, and electron tomogram. The thickness of section was 200 nm and HVEM images were obtained from about 500 serial sections. From all these images the distribution of microtubules, mitochondria, and nuclei was reconstructed as three-dimensional structure using IMOD program. The current data provide us more precise cellular information and better understanding on the animal vision mechanism in new dimension.

Synchronization of bursting rhythms in a population of neurosecretory cells in the silkmoth

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The brain of the silkmoth *Bombyx mori* has four pairs of neurosecretory cells producing an insulin-related peptide, bombyxin. Individual bombyxin producing cells in a pupa had an intrinsic, ultradian bursting rhythm and collective burst patterns showed a complex dynamics including spontaneous synchronization and desynchronization of different combinations of cells. Statistical cross correlation analyses of the burst rhythms revealed that one cell usually relate more closely to a few particular cells with a similar periodicity. Close investigation of phase differences between individual bursts of the related cell pairs revealed that an in-phase synchronous state was usually maintained for many cycles, while an anti-phase state was transient and lasted only one or two cycles, thereby suggesting that an in-phase state is significantly stable. On the other hand, in a particular pupa whose brain-retrocerebral neurohemal complex was completely isolated from remaining nervous systems to sever the connections with a heartbeat reversal mechanism, an anti-phase relationship was also observed between several cell pairs. The result suggests that a periodic signal from the heartbeat reversal mechanism plays a significant role in the phase synchronization dynamics of the coupled oscillator system. I propose a dynamic model for the system.

Annexin IX participates in the determination of timing of PCD

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"Anterior silk gland (ASG) of the silkworm, *Bombyx mori*, undergoes programmed cell death (PCD) during pupal metamorphosis, and the PCD is triggered by 20-hydroxyecdysone (20E). To identify the 20E-inducible genes that are involved in the PCD, we performed subtraction PCR using the ASGs incubated with or without 20E *in vitro*. We obtained seven novel genes, one of which was annexin IX. Annexins are Ca^{2+} and phospholipids binding proteins conserved throughout animals, but they have not been studied in detail. In the cultured ASGs, 20E dose-dependently up-regulated the annexin IX gene. By contrast, developmental profile of its expression in the fifth instar showed that annexin IX mRNA level was high when the ecdysteroid titer was low, and decreased with an increase in the titer. The expression was mostly ceased when the ASGs began the PCD sequence in response to ecdysteroid. The expression profile of annexin IX was different between tissues depending on whether they undergo PCD or not. In addition, tissues destined to die initiated the PCD sequence after annexin IX was down-regulated. These results suggest that annexin IX participates in the determination of timing of PCD by delaying or suppressing the execution of PCD.
"

What triggers diapause in the blue-banded bee?

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A breeding program for blue-banded bees (*Amegilla cingulata*) is currently under development. This will allow the use of these bees as pollinators in greenhouses in Australia, in particular for tomato pollination. Tomato growers wish to use *Amegilla* year round, which makes it is necessary to prevent the brood from entering diapause. We report on an investigation of what triggers diapause. Blue-banded bees have a short generation time and several generations are produced during spring and summer. Adults die in autumn and winter, and the brood hibernates as large larvae in their dark cells. This suggests that the onset of diapause is triggered either directly by the effect of lowered autumn temperature on brood development, or indirectly by the effect of light and/or temperature on the reproducing mother, who then hormonally triggers diapause in her offspring. We investigate what triggers diapause in two ways: A) Correlatively by:- Reviewing capture dates from collections at different locations and latitudes- Documenting bee activity at different locations and latitudesB) Experimentally by presenting females and brood with:- Elevated temperatures- Artificially increased day lengthThe results of this ongoing research will be reported.

Drought responsive genes in the sleeping chironomid, *Polypedilum vanderplanki*

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Organisms have evolved various mechanisms for adaptation to adverse environmental conditions. The sleeping chironomid, *Polypedilum vanderplanki,*is the largest multicellular animal known to tolerate almost complete dehydration without ill effect. The anhydrobiotic (cryptobiotic) larvae could survive at extreme temperatures from -270°C to +106°C (Hinton, 1968). We have demonstrated that rapid accumulation of trehalose coincided with loss of water form the larvae, also that cerebral regulation was not involved in the process of anhydrobiosis like plants or unicellular organisms. To address molecular events during desiccation going into anhydrobiosis in this insect, we cloned several genes concerning trehalose metabolism, water transport and so on, by RT-PCR using degenerated primers and screening EST database of *P. vanderplanki*, and subsequently carried out Northern blot analysis to measure amounts of expressions of these genes during desiccation. Consequently, we obtained five drought responsive genes at least. We will discuss how these drought responsive genes orchestrate anhydrobiosis in *P. vanderplanki*.

Effect of temperature on development & reproduction of the emma cricket, *Teleogryllus emma* (Orthoptera : Gryllidae)

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To establish the indoor-rearing system of the emma cricket, *Teleogryllus emma*, the effects of temperature on development and reproduction of the emma cricket have been studied. The influence of temperature on developmental periods of emma cricket was investigated under the eight temperatures of 15, 18, 21, 25, 27, 29, 31 and 35°C, the humidity of 60±5%R.H., and the 16L·8D photo regimen. The developmental periods of *T. emma* nymphs had a range of 124.8 days to 44.4 days at the temperature of 21 and 35°C, respectively. At 15 and 18°C, however, all tested individuals died before emergence. The highest emergence rate was 86.7% at 27°C, but there were no statistically significant differences among the temperatures. The adult weight increased with increasing temperatures although the weight at 35°C was decreased. In addition, the influence of temperature on reproduction of emma cricket was investigated under three temperatures 22, 25 and 28°C, the humidity of 60±5%R.H., and the 16L·8D photo regimen. The longevity of female/male adults were 65.8/79.2, 68.5/67.8, 46.8/57.4 days at the temperature 22, 25 and 28°C, respectively. The pre-oviposition and oviposition periods were 32.5 and 26.9 days at 22°C, 22.9 and 34.1 days at 25°C and 22.1 and 21.8 days at 28°C. The highest average fecundity per female was 737.3 at 25°C. As a result, the optimum of temperature for the development and reproduction of *T. emma* were 27~31°C and 25°C, respectively.

Oosorption in the stink bug, *Plautia crossota stali*: vitellogenin dynamics & the site of protein degradation.

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Oosorption is a phenomenon in which oocytes in the ovary are resorbed in response to internal and/or environmental factors. To understand its underlying physiological mechanism, the fate of vitellogenin and proteinase activity during oosorption in the stink bug, *Plautia crossota stali* were followed. The ovaries undergoing normal vitellogenesis rapidly accumulated vitellogenin as well as other proteins after adult emergence. Western blotting using an anti-vitellogenin antiserum showed that vitellogenin consists of several peptide fragments and is modified to form vitellin. When oosorption was induced by starvation, vitellogenin and protein contents in the ovaries began to decrease in 1 or 2 days. Western blotting of haemolymph samples from starving females failed to detect either ovary or oosorption-specific protein fragments. The results suggest that during oosorption vitellogenin/vitellin in the oocytes is rapidly degraded and released into the haemolymph as amino acids or small peptides that can no longer be recognized by the antiserum. Film *in situ* zymography revealed a high level of proteinase activity was not found in the degenerating oocytes but in the follicle cell layer in the ovaries.

Juvenile hormone induction of glutathione S-Transferase activity in the larval fat body of the common cutworm, *Spodoptera litura* (Lepidoptera: Noctuidae)

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The effect of juvenile hormone (JH) on the activity of glutatione S-transferase (GST) in the fat body of larval common cutwrom, ***Spodoptera litura*****, has been investigated. The activity of fat body GST in the newly ecdysed (day-0) 6th instar female larvae dramatically increased within 12 h by treating a single topical application of 100 µg pyriproxyfen (a juvenile hormone analog, JHA) in comparison with the untreated larvae, and a similar result was obtained in the treatment with JH III as well. In contrast, both 1- and 2-day-old larval fat bodies did not show significant increase on the GST activity in response to the JHA treatment. Furthermore, both amanitin and cycloheximide suppressed the JH-induction of the GST activity, suggesting that it was affected on the gene transcription level. In this study, we first time demonstrated that the GST activity in an insect fat body can be elevated on the gene expression by JH.**

BDNF promotes development of AL neurons of *Bombyx mori in vitro*

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This study was conducted to investigate the effects of BDNF on neurite growth and survival rate of antennal lobe (AL) neurons in primary culture, and production and secretion of BDNF in brain and retrocerebral complex of the silk moth, *Bombyx mori*. BDNF induced a significant neurite extension of some projection neurons in antennal lobe (AL) and also stimulated to significantly outgrow primary branches of principal neurites of interneurons. In primary culture with BDNF, 20-HE and 5-HT, AL neurons showed higher survival rate by BDNF and especially the highest by a combination of BDNF and 5-HT. Results from western blots, ELISA assay and immunolabelings suggested that BDNF is synthesized by median and lateral neurosecretory cells with molecular weight of 38 kDa, then transported to corpora allata, and finally perhaps secreted into hemolymph for its action.

Pattern of neuronal apoptosis in insect brain induced by 20-hydroxyecdysone

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This study was conducted to investigate a pattern of apoptotic neurons in brains of the silk moth *Bombyx mori* during metamorphosis and also determine whether 20-hydroxyecdysone (20E) induces brain neuron apoptosis *in vivo* and *in vitro*. Postembryonic brains were treated by TUNEL assay to detect apoptotic neurons. The 20E was injected into 3rd instar larva and then isolated brains were treated by TUNEL assay. To demonstrate whether apoptosis of cultured neurons are induced by 20E *in vitro*, dissociated pupal brain neurons were cultured with **2**0E and then treated by TUNEL assay. Apoptotic neurons were first found in brain of 1st instar larva. Brain neuronal apoptosis increased during 2nd and 3rd instar larval periods. Brains of late 4th instar larvae showed the most increased number of apoptotic neurons of all postembryonic periods and brains of late 5th instar larvae also exhibited a number of apoptotic neurons. Apoptotic brain neurons reduced in number in early wandering stage, but number of apoptotic brain neurons strikingly increased again in late wandering stages. Early pupae showed a gradual decreasing slope of apoptotic neurons, but in mid stage, pupae exhibited a new peak of apoptotic neurons. Injection of 20E into larva or pupa and culture of brain neurons with 20E resulted in increase of apoptotic neurons, suggesting that 20-HE might induce neuronal apoptosis in developing brains.

The neuropeptide, [His^7]-corazonin induces phase-related morphological changes in the desert locust, *Schistocerca gregaria*.

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A neuropeptide, [His^7]-corazonin, was isolated as the dark-color inducing neurohormone in locusts, *Locusta migratoria* and *Schistocerca gregaria*. In *L. migratoria,* it also induces morphometric changes related to phase polyphenism when injected into isolated-reared (solitary) nymphs. The present study aimed to examine the role of [His^7]-corazonin in the control of phase-related morphogenesis in *S. gregaria.* Gregarious (crowed-reared) adults have smaller F/C (F = hind femur length / C = head width) and larger E/F (E = forewing length /F = hind femur length) ratios than the solitary counterparts. Injection of [His^7]-corazonin (1 nmol) into solitary nymphs at the 2nd and 3rd instars caused a significant shift in F/C and E/F ratios toward the value typical for gregarious adults in both. In this case, it is important to analyze the results for locusts with 5 and 6 nymphal instars separately. A single injection of the peptide (2 nmol) into solitary nymphs at different instars indicated that the earlier the injection the larger the 'gregarizing' effects of the peptide. A similar effect was found on the number of antennal olfactory (basiconic type A and coeloconic) sensilla. These results suggest that [His^7]-corazonin plays an important role in the control of phase polyphenism in locusts.

Endocrine mechanisms on the termination of larval diapause in the bamboo borer (*Omphisa fuscidentalis* Hampson)

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During the diapause of the bamboo borer larvae, the ecdysteroid titer is very low and prothoracic glands (PG) exhibit low secretory activity. An application of JH analogue (JHA) induced pupation with an activation of the PG. To elucidate the hormonal mechanisms of the PG stimulation by JHA, we first examined the minimum JHA-stimulation to activate the PG. JHA-treated PG were transplanted to non-treated (recipient) larvae on successive day after JHA treatment, and the recipients were observed. Results showed that one-day stimulation was enough to induce pupation. The PG's activity of the recipients increased transiently 16 days after the transplantation after which hemolymph ecdysteroid increased gradually. In order to study the EcR mRNA expression, Omphisa EcR was cloned. The amplified region of Omphisa EcR was 973 base pair with high homology of the nucleotide of Junonia EcR (81.1%) and Manduca EcR (80.9%). Quantitative PCR was analysed to compare the EcR mRNA levels between JHA-treated PG and non-treated PG. The results showed that the expression of Omphisa EcR mRNA in JHA-treated PG was higher than in non-treated PG. From these results, it is suggested that JHA is tightly involved in the termination of the larval diapause in the bamboo borer by increasing the ecdysteroid titer and EcR mRNA expression in the prothoracic glands.

Postembryonic development of the wing imaginal discs in the female of the wingless-legged bagworm moth *Bacotia sakabei* (Lepidoptera: Psychidae)

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Wing reduction in female moths exists in several grades of the Psychidae; thus, studies on the wing degeneration process can help us understand the evolution of this family. In the Bagworm moth, *Bacotia sakabei*, the female adult has atrophied wings. However, the female pupa has vestigial wings. Using light and electron microscopy, I examined the processes of wing development during the last larval-instar stage and wing degeneration during the pupal stage in this species. Female wing imaginal discs proliferated during the last larval-instar stage. In the prepupal stages, however, wing imaginal discs degenerated due to programmed cell death (PCD). In the pupal stage, phagocytotic hemocytes floated within the bilayer of the wing epithelial cells in the female. This is also considered an apoptotic event, or PCD. Tracheae, which insert adult wing veins in the late prepupal stage, were undeveloped in the pupal wings. In the present study, I found that female-specific wing degeneration in *B. sakabei,* caused by programmed cell death, occurred two times, i.e. in the larval and pupal stages. Such a process of wing degeneration is probably unique to the bagworm moth of the family Psychidae.

Investigations on the fat body role in the dynamics of sunn pest populations

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In the sunn pest damaging area from Romania, three Eurygaster *species with different proportion, are present:* E. integriceps *(95 %),* E. maura *(3 %) and* E. austriaca *(2 %). The main factor which determines the population proportion is the fertility (*E. integriceps, *64 egg/female;* E. maura, *42;* E. austriaca, *29). This depends by the fat body value, very different between species. Detailed analyses on* E. integriceps *were performed. The accumulation of reserve matter and the fat body level present a great ununiformity, both individually and from one zone to another or from one generation to another, being the consequence of a complex of factors from which the climatic and agrotechnical ones are the most important. The fat body presents a significant role into life-cycle of this species, constituting one of the most important factors of perpetuation and numerical blasting with invasion role. The fat body level influences the mortality during diapause, the sterility and fertility, strongly influencing the bug population multiplication. The fat body is consumed in a proportion of 25% for maturation during diapause and 50% for oviposition. The multiplication index ranges between 0 and 56.47 depending on the fat body level.*

Factors in mating signal transduction in the ring-legged earwig.

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Mating is critical for the expression of oviposition and maternal care in earwigs; additionally, mating diminishes mating receptivity, and promotes a decline in juvenile hormone synthesis at the end of the gonadotrophic cycle. Severance of the ventral nerve cord of newly emerged, virgin females similarly promoted egg deposition and ensuing maternal care, diminished mating receptivity and elicited a timely decline in juvenile hormone biosynthesis. Mating of intact females to males castrated on day 1 of adult life resulted in reduced clutch sizes, but did not abolish fertilization; indeed, castrated males could produce viable offspring after 6 matings. Such castrated males had smaller seminal vesicles than did intact males, presumably attributable to sperm depletion. Mating of intact females to adult males that were castrated as larvae did not abolish oviposition; however, clutch sizes were reduced, and no eggs developed, indicating (not surprisingly) lack of fertilization. These results are consistent with the notion that ventral nerve cord severance mimicked mating. Following mating, the ventral nerve cord likely is a conduit to release the brain from inhibiting ovipositional and maternal behaviors. The presence of sperm in the spermatheca is not necessary for release of this inhibition, but may modulate clutch size.

Structure & function of sperm storage organs in **female** *Archisepsis diversiformis* **(Diptera: Sepsidae)**

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Female *Archisepsis diversiformis* has two spherical spermathecae, one larger than the other, and one ventral receptacle. The spermathecae are sclerotized, porous capsules surrounded by possibly glandular cells; part of the spermathecae capsule projects internally at the opening. The ventral receptacle has two non-sclerotized capsules and an expandable pedicel, all surrounded by muscle fibers. The pedicel, lined internally by spicules, has a chitinous plate and a circle or arch with spines. The spermathecae are for long-term storage of sperm, while the ventral receptacle is for short-term storage during fertilization. The spermathecae are filled 2-4 hours after a female's first mating. The sperm probably coil around the internal projection of the spermatheca, resulting in a organized, circular distribution of sperm. The small spermatheca is apparently the first to release sperm during oviposition, when the sperm are probably transferred to the ventral receptacle and stored temporarily in the two capsules. During the first 20 minutes of a second mating, the amount of sperm in the small spermatheca increases, while the amount of the large one decrease. Perhaps the large spermatheca re-fills the small one before the sperm of the second male are stored.

Early polytrophic meroistic oogenesis in the scorpion fly (Panorpa: Mecoptera)

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Scorpion flies (Genus *Panorpa*) have polytrophic meroistic ovaries. In *Panorpa*, an ovarian follicle consists of an oocyte, three sibling nurse cells and a monolayer of somatic follicle cells which encases the germ cell cluster. Germ cell clusters are formed in the germarium. A cystoblast runs through two successive rounds of synchronous mitoses followed by incomplete cytokineses. Thus, the four resulting cystocytes remain interconnected by three permanent ring canals. Using immunocytochemical assays we were able to detect significant differences between early oogenesis in *Panorpa* and the polytrophic model insect *Drosophila*. *Panorpa* germaria miss a distinct stem cell nice of the *Drosophila* type. The anterior compartment of the *Panorpa* germarium is occupied by a population of mitotically active, completely dividing germ cells, which represent the precursors of the cystoblasts. Neither by anti α-spectrin nor by phalloidin labelling we were able to identify spectrosomes in these cells. Dividing cystocytes accumulate α-spectrin and f-actin in their ring canals, not in a continuous polyfusome. In anti α-spectrin-labelled specimen a polyfusome becomes first visible after completion of cluster divisions. Subsequently the polyfusome is replaced by a polarised microtubule cytoskeleton. Ongoing ultrastructural studies will shed further light on the cellular processes underlying early oogenesis in *Panorpa*.

Gene & functional analyses of enzymes for juvenile hormone degradation

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Juvenile hormone (JH) esterase and JH epoxide hydrolase (JHEH) are known to regulate JH concentration in lepidopteran insects. First, cDNAs coding the JH esterase and JHEH of silkworm, *Bombyx mori* were cloned and sequenced, and mRNA expression was investigated by quantitative PCR. At mid-ultimate larval stage, almost all organs investigated were detected, whereas expressions were varied in organs at mid-penultimate larval stage. According to the results of developmental changes in mRNA expression and enzymatic activity of those enzymes, JH in larval body is cleared totally from mid-ultimate instar by hemolymph JH esterase and JH titer in each organs are regulated by cellular JH esterase and JHEH before the stage. Both JHEH and JH esterase have played also important role in embryos, and the rate of hatching fell by injection of dsRNA of JHEH into freshly laid egg in dose-dependent manner. Development of the individuals which injected with dsRNA of JHEH was abnormal when the embryo was observing after 5 day of the injection. It is speculated that the JHEH at embryonic stage controls JH concentration for every organs individually like at larval stages before last molting, and that high concentration of JH at certain organ(s) interferes normal embryonic growth.

Correlation of oxygen consumption rate, cytochrome C oxidase activity & CO I gene expression in larval diapause of the bamboo borer (*Omphisa fuscidentalis* Hampson)

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Bamboo borer is a larva of the moth, Omphisa fuscidentalis *Hampson (Lepidoptera, Pyralidae). Previous study shows that juvenile hormone analogue (JHA, S-methoprene) induces pupation of the diapause larvae with increasing the ecdysteroid titer through the prothoracic gland activation. At the time of diapause termination, metabolic rate of the diapause larvae may increase due to an activation of cellular respiration by ecdysteroid. Accordingly we studied the changes in oxygen consumption rate with the correlation to cytochrome C oxidase activity and CO I gene activation. Results indicated that oxygen consumption rate showed a supradian rhythm, at about 12 hrs. cycles at 25°C during diapause. During the 6-month larval diapause from October to March, oxygen consumption rate was significantly higher in March than in October to January ($P<0.05$). When 1 mg JHA was topically applied to diapause larvae, rate of oxygen consumption increased 7 days after the application. The rate decreased slightly in G0, increased in G1-G2 and then reached a maximum at pupation. Next we studied how the cytochrome C oxidase activity changes after application of JHA. Cytochrome C oxidase activity increased gradually after 1 day of JHA application and reached the maximum level at pupation ($P<0.05$). These indicating that termination of diapause, either it occurs naturally or artificially induced by JHA, associates with increasing of oxygen consumption rate and cytochrome C oxidase activity. JHA may elicit a high metabolic activity that is correlated with the increasing of cytochrome C oxidase activity and oxygen consumption rate in the larval-pupal transformation. A partial sequence of cytochrome C oxidase subunit I (CO-I) was used to examine the CO-I mRNA expression in the diapause larvae mid gut following the JHA application. Results of semi-quantitative PCR showed that CO-I mRNA increased gradually from day 4 of JHA application to the pupation. These results suggests that JHA is tightly involved in the termination of the larval diapause in the bamboo borer by increasing the ecdysteroid titer, which causes an increase in oxygen consumption through the activation of CO-I gene and cytochrome C oxidase activity.*

Localization of allatostatin & FMRFamide in brain & retrocerebral complex of the termite *Reticulitermes flavipes* & the effect of allatostatin on juvenile hormone synthesis

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A family of allatostatins with the C-terminus Phe-Gly-Leu amide was discovered in a cockroach by the inhibition of juvenile hormone (JH) synthesis by the corpora allata (CA). These peptides occur throughout the nervous system in many groups of insects and also have many other functions. The effect on CA has, to date, been shown only for cockroaches and crickets in which axons from lateral neurosecretory cells of the protocerebrum arborize extensively in the CA. Some of these cells show allatostatin colocalized with FMRFamide in the cockroach *Diplptera punctata.* Because termites and cockroaches are closely related we have investigated the localization of cockroach-like allatostatin and FMRFamide-like peptides in different casts of the termite *Reticulitermes flavipes.* Both medial and lateral neurosecretory cells of the protocerebrum are immunopositive for cockroach-like-allatostatin and a few of these cells show colocalization of FMRFamide-like immunoreactivity. Extensive arborization of allatostatin immunoreactive axons occurs in the CA of secondary reproductives but less is found in the CA of workers and nymphs. *D. punctata* allatostatins inhibit JH synthesis by CA of secondary reproductives in a dose dependant way. Thus Phe-Gly-Leu amide allatostatins affect JH synthesis in this termite species and very likely in other termite species as well..

Regulation of prothoracic gland ecdysteroidogenic activity leading to pupal metamorphosis.

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Insect metamorphosis is under the controlled of ecdysteroid secreted from the prothoracic glands (PGs). PGs are activated by prothoracicotropic hormone (PTTH) and under the feedback regulation. Comparison of the gland activities in the third, fourth and fifth instars showed that PGs in early fifth (final) instar is unique: they neither exhibit ecdysteroid secretion nor respond to PTTH. We supposed that this 'silent' period was the first signal leading to the pupal metamorphosis. We examined a possible involvement of feedback control in the silence by examining the PGs activity after injection of 20-hydroxyecdysone (20E). 20E inhibited PG activity in the late fourth instar stronger than that in early fourth instar. When 1μg of 20E was injected in early fourth instar, we discovered the activity never appeared again. By contrast, the PGs of larvae whose brain-corpora allata complex had been removed were once inhibited by 20E but recovered the secretory activity. Combined data suggest that the sensibility of the PGs to 20E increases through the penultimate instar, and JH suppresses the recovery from the feedback inhibition. We also determined the developmental profiles of EcR and USP in order to identify the factor(s) involved in the changes of the sensibility to 20E.

Effects of photoperiod & temperature on diapause development in the nymph of the scutellerid bug, *Poecilocoris lewisi*

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Poecilocoris lewisi (Heteroptera: Scutelleridae) enters diapause in the 5th (final) nymphal instar under short-day conditions. The critical daylength inducing this nymphal diapause is 14.5 h at 25°C, and this photoperiodism causes a partially bivoltine life cycle in central Japan. When diapause nymphs were transferred to longer daylengths, they terminated diapause and emerged as adults. Diapause was terminated after a long period even under the same conditions that induced the diapause. Low temperature treatment of 10°C had only a little effect on the diapause termination. Transfers of diapause nymphs from natural conditions to the laboratory conditions revealed that the diapause development proceeds gradually but not constantly from autumn to spring. As a result, the nymphal diapause was maintained until late February under natural conditions. The diapause development, however, had been completed in late December under natural photoperiod at constant 25°C. We suggest that in nature the diapause development proceeds constantly during the temperature is relatively high, and after the temperature becomes low, the rate of diapause development decreases. The effect of temperature on diapause development is regarded as a mechanism to keep both the first and the second generations in diapause until early spring.

Effects of 20-Hydroxyecdysone & juvenile hormone analogue on the development of wing imaginal disks of the bamboo borer, *Omphisa fuscidentalis* Hampson

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Larvae of the bamboo borer, *Omphisa fuscidentalis* are in diapause for nine months, from September until the following June. To examine the growth and differentiation of wing imaginal disks, size measurement and protein assay of disk were determined during September to April. Results showed that both fore wings and hind wings size of diapausing larvae increased gradually from October to April. Pupal induction of diapause larvae by different concentrations of 20-hydroxyecdysone(20E) and JHA application caused the wing imaginal disk growth and differentiation in dose-dependent manner. Previous studies show that topical application of JHA induces pupation by increasing of ecdysteroid titer through the prothoracic glands activation. The expression of OfEcR-B1 in the wing disks was measured by semi-quantitative PCR after JHA application. Results showed that OfEcR-B1 mRNA expression in the wing imaginal disks increased from day 5 to day 10 of JHA application and rapidly decreased at pupation. These results suggest that JHA terminated the larval diapause of the bamboo borer by increasing the ecdysteroid titer which induced the EcR-mRNA expression and the development of wing imaginal disks.

An involvement of juvenile hormone analogue & ecdysone receptors on changes of trehalase activity of the bamboo borer, *Omphisa fuscidentalis* *Hampson.*

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During larval diapause from September to May trehalose concentration in hemolymph remains between 40-50mM and decreases to an almost undetectable level after pupation. Since trehalose metabolism, depend on trehalase locating on surface of midgut, trehalase activity in midgut homogenates was measured through the larval diapause period and pupal stage. The midgut homogenate exhibited low trehalase activity from December to April, showed a 4-folds increase in May and remained high at pupal stage in July. After JHA application, trehalase activity increase gradually attained the maximal level at pupal stage. Injection of 20-hydroxyecdysone (20E) caused an increase of trehalase activity in vitro and in vivo. To examine an involvement of 20E and JHA on the change of trehalase activity during larval diapause, two isoforms of ecdysone receptor from the bamboo borer,*Omphisa fuscidentalis* (OfEcR) were identified and examined their expression level in the midgut during diapause period and after JHA application. The expression of both isoforms in midgut was studied by RT-PCR. After JHA treatment, the expression level of EcR-A and EcR-B1 correlated with the time of increasing titer of hemolymph ecdysteroid. Suggest that JHA stimulated an increasing of hemolymph ecdysteroid, then ecdysteroid induced the transcription of EcR gene and trehalase activity.

Developmental profiles of two storage proteins from the bamboo borer, *Omphisa fuscidentalis.*

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Storage protein 1 (SP1) and storage protein 2 (SP2) are major components of haemolymph proteins of diapausing larvae of the bamboo borer, *Omphisa fuscidentalis*. They were purified from the larval haemolymph by ammonium sulfate precipitation, followed by three difference chromatographies using Q-sepharose, superose 6 and Mono Q columns. Each purified protein exhibited a single band on SDS-PAGE. The molecular sizes of SP1 and SP2 were estimated to be 75 and 72 kDa, respectively. Gel chromatographical analysis indicated that the molecular sizes of SP1 and SP2 in native conditions were 423 and 403 kDa, respectively, showing that these storage proteins were hexamers. Storage proteins in lepidopterans are usually synthesized by fat body and secreted into haemolymph in feeding period and reabsorbed into the fat body during prepupal period. We therefore examined the developmental profiles of SP1 and SP2 concentrations in male and female pupae by Western blot using rabbit polyclonal antibodies. The results showed that SP1 and SP2 concentration in haemolymph dramatically decreased while gradually increased in fat body during pupation. The study indicates that SP1 and SP2 are incorporated into fat body at the time of diapause break., like other storage proteins. Partial amino acid sequence of those proteins indicated that SP1 is highly homologous to arylphorin and SP2 to hexamerin.

Induction of cryptobiosis excised-fat body of the sleeping chironomid, *Polypedilum vanderplanki*

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A rain pool dweller, *Polypedilum vanderplanki*, can stand complete dehydration. This species is known as the only insect able to enter cryptobiosis. The larvae accumulated a large amount of trehalose during dehydration. We have demonstrated that induction of cryptobiosis occurred without the cerebral regulation in this chironomid. To confirm whether individual tissues can prepare for entering cryptobiosis by themselves, excised tissues were dehydrated *in vitro* with various kinds of media. Excised-fat body desiccated gradually for 2 days *in vitro* with BmHem (heat-treated hemolymph of *Bombyx mori* larvae) accumulated a large amount of trehalose and recovered even after extended dry preservation for 3 months. Excised-alimentary canal in contrast produced little trehalose *in vitro* during dehydration with the same medium and failed to recover after rehydration. Excised-fat body quickly desiccated within a day with BmHem contained only a small amount of trehalose and failed to recover. Based on these results in *P.vanderplanki*, we discuss the significant roles of trehalose to protect tissues and cells of the cryptobiotes during complete desiccation.

Termination of reproductive diapause in adults of the rice bug, *Leptocorisa chinensis*

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The rice bug, *Leptocorisa chinensis*, is one of the major insect pests of rice in the south to the central Japan. Field studies have revealed that *L. chinensis* spend two generations a year in Tsukuba (36°01'N, 140°06'E). Overwintering adults appear on gramineous weeds in late June to early July and two successive generations occur during mid July to late September. Adults of the second generation enter reproductive diapause after mid September. To specify the timing of diapause termination, second generation adults were collected late in October and kept in cages under outdoor conditions. These insects were transferred monthly from the cage to laboratory conditions in January to June, and reared as single male and female pairs under a long-day (16L8D, LD) or a short-day (12L12D, SD) condition at 25°C. SD maintained reproductive diapause and LD gradually shortened the pre-oviposition period in females transferred from cages between January and late May. Females transferred from cages in mid June showed a short pre-oviposition period under both LD and SD. The results show that the long day length is necessary for the diapause termination in *L. chinensis*.

Effects of locomotive phase on reproduction in the German cockroach (*Blattella germanica*) & the double-striped cockroach (*B. bisignata*)

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The German cockroach (*Blattella germanica*) and the double-stripped cockroach (*B. bisignata*) are sibling species, but their habitats are dramatically different. The former is a cosmopolitan, household species, whereas the latter is a complete feral species and distributed only in Southeast Asia. It has been demonstrated that the mate-finding locomotion of both species showed circadian rhythmicity. Apparently, the circadian clock is involved in the reproductive behaviors. Since the mate-finding locomotion represents the calling behavior, which is the first step of the mating sequence, the courtship and copulation are the following steps leading to a successful mating. The contribution of circadian timing on the success of each mating sequence is investigated in detail.

Thermoperiodic restoration of *Drosophila* circadian rhythms in constant light

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Although circadian locomotor rhythms of the fruit fly *Drosophila melanogaster* disappear in constant light (LL) under constant temperature, the rhythm is restored by exposing to temperature cycles. We examined molecular and cellular basis of this thermoperiodic restoration of the rhythm. Several transient cycles were required to establish steady state entrainment, suggesting that the rhythm is driven by an endogenous circadian clock. PER and TIM exhibited rhythmic expression in their abundance under temperature cycles, both increasing in cryophase and decreasing in thermophase. Arrhythmic mutant flies, *per*^{01} and *tim*^{01}, also showed rhythmic locomotor activity under temperature cycles in LL, but the rhythm was quite different from that of wild-type flies. *disco*^{2} mutant flies that lack lateral neurons (LNs), putative clock neurons, also show abnormal rhythmicity similar to that of *per*^{01} and *tim*^{01} flies. These results suggest that rhythmic expression of *per* and *tim* is involved in the restoration of the locomotor rhythm by temperature cycles and that LNs play an important role in this rhythm restoration.

Interaction of Juvenogens and Insect Oostatic Peptides with Developing Ovaries of *Neobellieria bullata* (Diptera)

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Juvenogens, fatty acid esters of selected insect juvenile hormone bioanalogues, and insect oostatic peptides (H-YDPAP-OH and H-YDPA-OH) were selected from the screening programme on a model object *Neobellieria bullata* (Diptera) as the most promising ones. They were applied by injections (peptides) or by topical application (juvenogens). At given intervals the flies were dissected and their ovaries were examined for morphological changes and hatchability of eggs in the uterus, respectively. Under described regime of applications the development of the first egg was not morphologically affected. Hatchability of eggs in uterus slightly decreased, ranging from 60 to 80 percent compared to controls. The second gonotrophic cycle was characterized by remarkable changes in the follicular epithelium of about 60% of oocytes. Monolayered cylindrical follicular epithelium with nuclei in the centre is altered the following way: first the nucleoli became prominent and subsequently multiplied, nuclei show slight difference in their size. Some bizarre nuclei with great number of nucleoli start to appear; nucleoli often merge together. The nucleocytoplasmatic ratio grows for the benefit of nucleus. Groups of giant cells appear. The epithelium proliferates towards the inner part of egg chamber. In some sections the follicular epithelium forms pseudopapillar projections, stratification is severely disturbed and extensive invagination to the centre of the egg chamber appears.

Making many, few, or no eggs: The structural and molecular basis of oogenesis in polyphenic social bees

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Plasticity in reproduction is a hallmark of social insects. To understand the underlying mechanisms we investigated the structural organization of queen and worker ovarioles of two highly eusocial bees. In the honey bee, we observed clusters of potential stem cells in the terminal filament. Mitotic activity was confirmed by BrdU incorporation, and their germ line nature is currently being checked by *vasa* expression. Microfilaments and microtubule organization analyzed by immunofluorescence microscopy revealed progressive oogenesis in the germaria even in workers kept in the presence of the queen. The process of vitellogenesis was probed by Northern Blot and RT-PCR analysis of vitellogenin expression in the fat body and in the ovaries, and its regulatory circuitry was investigated by RNA interference followed by juvenile hormone titer analysis. These studies revealed a negative feedback relationship between vitellogenin expression and the hormone titer in worker bees, adding an important facet to the mechanisms underlying reproduction and division of labor. In the stingless bee, the ovarioles of newly ecloded queens and workers contained one follicle undergoing previtellogenic growth, thus deviating considerably from the picture observed in the honey bee. The observed levels of plasticity can be related to life history strategies in these bees.

Thermoperiodic restoration of *Drosophila* circadian rhythms in constant light

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Although circadian locomotor rhythms of the fruit fly *Drosophila melanogaster* disappear in constant light (LL) under constant temperature, the rhythm is restored by exposing to temperature cycles. We examined molecular and cellular basis of this thermoperiodic restoration of the rhythm. Several transient cycles were required to establish steady state entrainment, suggesting that the rhythm is driven by an endogenous circadian clock. PER and TIM exhibited rhythmic expression in their abundance under temperature cycles, both increasing in cryophase and decreasing in thermophase. Arrhythmic mutant flies, *per*^{01} and *tim*^{01}, also showed rhythmic locomotor activity under temperature cycles in LL, but the rhythm was quite different from that of wild-type flies. *disco*^{2} mutant flies that lack lateral neurons (LNs), putative clock neurons, also show abnormal rhythmicity similar to that of *per*^{01} and *tim*^{01} flies. These results suggest that rhythmic expression of *per* and *tim* is involved in the restoration of the locomotor rhythm by temperature cycles and that LNs play an important role in this rhythm restoration.

Synaptogenesis in the giant fibre system of *Drosophila*

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Electrical synapses, which allow direct signal transmission are, in effect, gap junctions. In invertebrates, innexins are the major structural components of gap-junction channels (Phelan et al 98, Nature 391: 181-184). The *shaking-B* locus encodes three members of this family: ShakB(neural), ShakB(N+16) and ShakB(lethal). Here, we show that electrical synapse formation between the Giant Fibre (GF) and its major synaptic target, the tergotrochanteral motor neuron (TTMn), requires expression of different innexins on each side of the synapse. The mutation *shaking-B*^{2}, which removes both ShakB(neural) and ShakB(N+16) function, disrupts dye coupling between the GF and TTMn (Phelan et al 96, J Neurosci 16: 1101-1113), although the two neurons still come into cell-cell contact in this mutant (Blagburn et al 99, J Comp Neurol 404: 449-458). Expression of ShakB(N+16) in the GF and TTMn is sufficient to restore dye coupling in the mutant. However, expression of ShakB(neural), identical to ShakB(N+16) but for the absence of the first 16 amino acids, fails to afford rescue. Expression of a dsRNA construct against ShakB(lethal) in the TTMn abolishes dye coupling between the GF and TTMn. Thus, expression of ShakB(N+16) in the GF and ShakB(lethal) in the TTMn are required for electrical synapse formation between these neurons.

A heavy load of sperm; male investment in reproduction of *Yponomeuta* moths.

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Females are choosy, as is generally believed, because of their large investment in reproduction compared to males. However, male investment can be considerable. Males in two closely related moth species, *Y. padellus* and *Y. cagnagellus*, lose on average respectively 13.5% and 17.2% of their body weight during spermatophore transfer to the female. Therefore, they invest substantially into reproduction. It has been suggested that male investment should be positively correlated with sperm competition. This is currently investigated. *Y. cagnagellus* mates more often than *Y. padellus*, probably increasing the level of sperm competition. Therefore, *Y. cagnagellus* should invest more than *Y. padellus.* Indeed, we found that this species transfers a significantly larger percent of its bodyweight to the female. If higher sperm competition leads to maximization of sperm numbers, we expect that spermatophores of *Y. cagnagellus* to contain relatively more sperm than spermatophores of *Y. padellus.* Furthermore, it has been suggested that some male lepidopteran species can tailor their ejaculate according to sperm competition risks i.e. spermatophore weight and number of sperm transferred to the female can vary according to female mating status and body size. We will therefore investigate sperm transfer for first and second matings in the two *Yponomeuta* species.

Boundary between two cell types coordinates follicular morphogenesis

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Oogenesis in the fruit fly *Drosophila melanogaster* is an excellent model system for studying how cell-cell interactions regulate cell shapes and movements during development. In the ovary, follicle cells surround the oocyte in a single-layer sheet; signaling from the oocyte induces a subset of follicle cells to change their shape and position and form two tubes. These cells secrete eggshell proteins into the tube lumens to produce the dorso-lateral eggshell appendages, which facilitate respiration in the developing embryo. Interactions between cells coordinate this morphogenetic process. Two cell types make each tube: cells expressing the transcription factor Broad form the roof and sides of each tube, while cells expressing the *rhomboid-lacZ* reporter gene make the floor. A combination of signals involving EGF-receptor and BMP2/4 pathways specifies these cell types. The Notch pathway participates in the process that defines a border between the Broad- and *rho-lacZ*–expressing domains. The roof- and floor-forming cells express different cell-fate and adhesion markers, exhibit distinct behaviors, never intermingle, and coordinate their movements, even in patterning and morphogenesis mutants. We hypothesize that a boundary exists between these two cell types and that signaling across the boundary coordinates the cell shape-changes and rearrangements that form the tube.

The rhythmic component of photoperiodic time measurement: cause or consequence?

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Organisms at temperate latitudes encounter seasonal environments that vary with geography and modify their seasonal programming of development and diapause accordingly. The mosquito, *Wyeomyia smithii*, ranges from the Gulf of Mexico to northern Canada where it occupies only the water-filled leaves of a single species of pitcher plant. This constancy of habitat makes it ideal for studying the relationship between photoperiodism that regulates seasonal activities and the circadian clock that regulates daily activities. In *W. smithii*, the critical photoperiod mediating the switch from continuous development to diapause tracks latitude and altitude more closely (R^2>90%) than any other known ecogeographic trait. Concomitantly, the rhythmic response to the Nanda-Hamner protocol, historically used to infer a circadian basis for photoperiodism, declines with increasing latitude and altitude (R^2 = 70%). Critical photoperiod and the rhythmic response to Nanda-Hamner are genetically correlated within populations, but not between populations. Hence, photoperiodic response is capable of evolving independently of the circadian clock, even within a single species. The relationship between photoperiodism and circadian rhythmicity may be correlational but is not causal. Elucidating the mechanics of photoperiodisn will involve the genetics of photoperiodism itself, rather than considering it simply as an adjunct to the circadian clock.

Stem cells, germ cell cluster formation and fusomes in insect gonads

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Throughout the animal kingdom germ cells differentiate into oocytes and sperms. Oocytes are immobile and have to grow into giant cells, thereby providing all the resources needed for early embryogenesis. Sperms are mobile and a huge number is needed to ensure fertilization. As a consequence, sperms minimize their volume. This division of labor between gametes is achieved by sex-linked differentiation of germ cells. Both, the male and the female pathway start with a multiplication strategy. In the beginning, a few germ cells succeed in becoming stem cells which undergo differential mitoses. In doing so, cystoblasts are generated from which syncytial germ cell clusters arise by a number of synchronized mitoses coupled with incomplete cytokinesis. In most insects studies so far, this cluster formation is accompanied with a special germ cell organell, the fusome. The fusome is build and maintained during the cluster multiplication steps. These intrinsic steps of germ cell differentiation are coupled with extrinsic germ cell - somatic cell interactions. Variations of stem cell activity, as well as the variations during germ cell cluster development, both in combination with extrinsic factors, are sufficient to generate the differences leading to the panoistic, polytrophic and telotrophic organization of insect ovarioles.

How similar are daily and seasonal biological clocks?

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Daily and seasonal timing systems in insects have usually been supposed to share similar mechanisms, because both rely in large measure on information from the daily light-dark cycle: daily clocks can ensure that activity coincides with the appropriate time of day, and seasonal time is indicated most reliably by daylength. However, several lines of evidence suggest that the systems are different. For example, receptor features, photosensitive pigments, clocks, and the effectors that mediate responses to information derived from the clock may have different daily, seasonal and general functions and properties, and several different systems are known. Therefore, it is unlikely that there is any single master clock for insect timing, despite the long-standing belief that such a thing exists. The absence of a single clock would be consistent with the fact that temporal responses serve a very wide range of purposes, meeting many different ecological needs on different time frames.

Wigglesworth Memorial Lecture. From insect ovaries to sheep red blood cells: a tale of two hormones

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Insect juvenile hormone is widely acknowledged to act at least two levels in the cell. In exercising its metamorphic control, it is held to interact in some way with the genome. Certain other effects, such as those controlling the volume and shape changes of follicle cells during yolk uptake, are the result of JH interacting with a membrane receptor. The vertebrate hormone thyroxine exerts many of its effects via a well described nuclear receptor complex, but also has some more immediate effects through a yet to be fully characterised membrane receptor. Although the two hormones are very different in structure, they are, in their membrane effects, mimics of one another: some thyroid hormones act on the follicle cells as mimics of JH, and JH mimics the membrane effects of thyroxine on mammalian red blood cells. The evidence suggests that the connecting link may be a sensitivity of the membrane receptors to CO_2. The implications of these discoveries for the origin and evolution of receptors will be explored.

Genes involved in flesh fly diapause: lessons from microarrays

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One of the challenges of working with a non-genetic organism is the lack of available mutants and genomic databases for molecular analysis. This challenge is especially acute for work on insect diapause because none of the genetically-known organisms have a well defined diapause. Until such databases become available for species that have a diapause, an alternative is to use cross-species hybridization. In our case, flesh flies (*Sarcophaga crassipalpis*) and *Drosophila melanogaster* appear to be sufficiently close phylogenetically to allow us to captilalize on microarray chips available for *D. melanogaster.* This approach has enabled us to identify numerous genes that are uniquely up- or down-regulated during early, mid-, and late-diapause in *S. crassipalpis.* The most abundantly expressed genes associated with diapause are those encoding stress proteins and structural proteins associated with the cytoskeleton. Genes of potential regulatory importance include those that encode regulatory proteins, including transcription factors, and signal transduction proteins involved in the *shaggy, notch* and *wingless* pathways.

Comparative studies on germ cell formation in crustaceans and insects: patterns of epigenesis and preformation with respect to arthropod phylogeny

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The diversity in the temporal and spatial origin of arthropod germ cells is extreme. Studies on model organisms indicate that germ cells may be specified through the asymmetric localisation of germ line determinants (germ plasm), or by non-autonomous signals from neighbouring cells to germ cell precursors. In *Drosophila melanogaster*, germ plasm components are synthesised during oogenesis by nurse cells and transported to the oocyte, where they are then asymmetrically localised to the region of ooplasm that will form germ cells. However, this dipteran model cannot be applicable to all hexapods, since in many species of primitive insects, oogenesis functions without nurse cells; indeed, most of the basal insect orders for which data are available do not appear to have early segregated germ cells. Since recent arthropod phylogenies suggest that crustaceans and insects are sister groups, examination of both basal insects and crustacean taxa can help to determine what kinds of germ cell specification mechanisms might have been ancestral to insects. I have used a molecular marker to study germ cell development in the amphipod crustacean *Parhyale hawaiensis*, and have determined the capacity of individual early cleavage blastomeres to form germ cells by blastomere isolation and culture. My experiments provide experimental evidence that germ cell determinants are present in only one cell at the two-cell stage in *P. hawaiensis*. These results will be discussed within the context of the existing literature on crustacean and hexapod germ cell specification.

Sperm storage dynamics: when molecular techniques and morphological studies meet

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The tephritid fruit fly, *Anastrepha suspensa*, has a complex reproductive system which includes four sperm storage organs and offers many potential opportunities for selection on female-controlled sperm storage and use. Thus, this species provides a good model system for examining sperm partitioning patterns from multiple matings. Microsatellite primers and sperm amplification techniques were developed to examine sperm partitioning from multiple ejaculates in wild populations of this species to determine potential effects on paternity outcomes.

Sperm competitive ability in stalk-eyed flies is influenced by meiotic drive and seminal fluid

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Sperm competition is common in many insect species; however, the mechanisms underlying differences in sperm precedence are not well understood. In the stalk-eyed fly, *Cyrtodiopsis whitei* (Diptera, Diopsidae), sperm precedence is influenced by the presence of sex chromosome meiotic drive. When drive-carrying males compete with non-driving males for fertilizations within a female, the number of progeny sired by drive males is significantly fewer than predicted by sperm mixing alone. Thus, drive males apparently suffer not only a reduction in the number of viable sperm produced, but also a reduction in sperm competitive ability. By interrupting copulations before sperm, but after seminal fluid, was transferred, we found that seminal fluid from another male influences the number of progeny sired by a drive-carrying male when both males mate with the same female. Sperm viability staining revealed that sperm from drive males are incapacitated by seminal fluid from other males within the female reproductive tract. These results suggest that multiple mating by females enables seminal fluid products to interact differentially with sperm and may reduce the transmission advantage of the drive chromosome.

Significance of prolonged copulation in reproductive success of *Dysdercus koenigii*

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The adults of *Dysdercus koenigii* remain in continuous copula up to 72 hr. The adaptive significance of prolonged copulation in the reproductive bioactivities of *Dysdercus* was investigated by a series of interrupted mating experiments. The adults were allowed to mate for a specified duration. The studies indicated that insemination in the female takes place during the first day of mating. The female, if mated for a longer time interval acquired additional sperm and male accessory gland secretions which modify the reproductive behavior of the female. Duration of mating had a profound effect on onset of oviposition and total number of eggs and egg batches laid by the female in its life span. Duration of mating was correlated to fertility of the female. Therefore, a female if mated for 24hr laid lesser number of fertile eggs. The fertility was significantly decreased in the egg batches laid during later part of life. The mated female exhibited a rhythm in ovipositional behavior. For instance, the female mated for shorter time intervals exhibited a change in magnitude of egg laying and ovipositional rhythm. Duration of mating was found a crucial factor in governing oviposition behavior. Further, the rate of oocyte growth was dependent on duration of mating as evidenced by a shorter inter-oviposition period in case of females mated for longer duration.

Endocrine regulation of a diapause-associated increase in fat storage in flesh flies.

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The flesh fly, Sarcophaga crassipalpis, spends the winter as a diapausing pupa. To support metabolism during this long period of dormancy, diapause-destined larvae accumulate significantly greater lipid stores than direct developing larvae. We have shown that diapause destined larvae accumulate their greater lipid stores by an overall increase in anabolic growth that yields individuals that are both larger and fatter. We have also shown that diapause-destined larvae feed for significantly longer to support this increase in body size and lipid stores. Because insulin signaling is important for regulating growth and metabolism in both vertebrates and invertebrates, we cloned a probe for the Sarcophaga insulin receptor homologue and used it to quantify receptor expression in diapause and direct developing individuals during larval feeding, wandering, and pupal development. We discuss our findings with regard to the role of insulin signaling in generating increased growth and lipid storage in diapause-destined flesh fly larvae.

Ecdysone receptor function and structure

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We have cloned and characterised full-length cDNAs encoding both EcR and USP subunits for ecdysone receptors from the cotton bollworm *Helicoverpa armigera* (Lepidoptera), the Australian sheep blowfly *Lucilia cuprina* (Diptera) and three members of the Hemiptera, the peach aphid *Myzus persicae,* the silverleaf whitefly *Bemesia tabaci* and the green vegetable bug *Nezara viridula*. The cDNAs have been placed into expression vectors and transfected in to CHO and CV1 mammalian cells along with reporter genes functionally linked to ecdysteroid responsive promoters. The ability of the receptors to activate reporter genes in response to insect ecdysteroids, phytoecdysteroids and a commercial synthetic compound has been studied. The responses of our cloned ecdysone receptors to different ligands will be discussed. Full-length receptor encoding cDNAs have been expressed in an *in vitro* transcription and translation system to examine the protein products of their open reading frames. Regions from within both EcR and USP subunits of our cloned receptors have also been subcloned for expression in a baculovirus system. Studies of the properties of the expressed protein segments, including competition ligand binding *in vitro*, are in progress. The three-dimensional structure of the *B. tabaci* ecdysone receptor heterodimeric ligand-binding domain has been determined by X-ray diffraction.

Diapause and circadian clock gene expression in the linden bug, *Pyrrhocoris apterus*

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Although photoperiodism is undoubtedly one of the most important functions of the circadian system, the role of circadian clock genes remains unclear. Molecular cloning of the pivotal clock gene homologs *period* and *Clock* from the European linden bug, *Pyrrhocoris apterus*, enabled us to test their possible role in the bug photoperiodic timing system. We compared the expression of *period* and *Clock* genes in the head of animals kept under diapause promoting short days (SD) and diapause preventing long days (LD), using RNase protection assay. There was only a weak diurnal rhythm in both *period* and *Clock* mRNA under LD and no detectable rhythm under SD. Under SD, however, the level of *period* mRNA was about 10-fold, and that of *Clock* mRNA about 2-fold higher than under LD. In a mutant, that does not undergo diapause, even under SD, levels of both transcripts were low in both photoperiods. The block to diapause photo-responsiveness was not associated with defects in the circadian timing, but levels of *period* and *Clock* mRNAs were related to the properties of activity rhythms. The differential regulation of the levels of two clock gene transcripts in a photoperiodic mutant strongly indicates a link between photoperiod, the magnitude of clock gene expression, and developmental outputs.

Accessory nuclei as a vehicle for transporting snRNPs in hymenopteran oocytes

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In some insects, oocytes contain characteristic organelles termed accessory nuclei, which originate by budding off the oocyte nucleus and are surrounded by an envelope identical to the nuclear envelope. In the common wasp, *Vespula germanica*, within accessory nuclei dense inclusions, immersed in translucent ground substance, occur. Ultrastructural analysis revealed that these inclusions form gradually during oogenesis and consist of a spherical homogenous body with an attached hemispherical granular structure. EM immunogold labelling and in situ hybridization experiments demonstrated that the mature inclusions contain small nuclear RNAs and proteins: Sm, coilin and SMN, factors involved in pre-mRNA splicing. In the investigated species, in early previtellogenic oocytes, the accessory nuclei surround the oocyte nucleus but as oogenesis progresses, they migrate to the cortical ooplasm and remain there until early embryonic development. Together, these results suggest that the accessory nuclei may be involved in transporting and localizing small nuclear ribonucleoproteins (snRNPs) to the oocyte periphery.

Fertilization chamber and ventral receptacle: diversity in form and function of a homologous structure in the female reproductive tract of higher flies

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In addition to 1-4 spermathecae, which are always connected to the dorsal vagina wall, a further sperm receptacle, the ventral receptacle (VR), occurs in most acalyptrate flies. The VR is positioned in the ventral vagina wall directly opposite to the spermathecal ducts. It functions as a temporary reservoir of sperm. In several instances the VR was found to be the fertilization site. Moreover, in some taxa it has completely taken over the function of sperm storage from the spermathecae. Conformity of position and function support the homology of the VR with the fertilization chamber, known to occur in several calyptrate flies. However, the VR is generally much more elaborate, varying from one-chambered to tubular or multi-chambered, from membranous to strongly sclerotized, from an inconspicuous pouch to a large and sometimes bizarre vessel. Because the VR holds a key position for fertilization, its diversity in shape and size must have implications for the possibilities of sperm competition and female sperm selection. New results about the morphology of the VR in various families will be presented along with hypotheses for its evolution from a simple fertilization chamber to the various elaborate shapes and sizes.

Development of the telotrophic meroistic ovary of the bug *Dysdercus intermedius*

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Bug ovaries are of the telotrophic meroistic type. Nurse cells are restricted to the apical tropharium and are in syncytial connection with the oocytes via cytoplasmic projections. It is unknown whether the syncytium is established by cell divisions with incomplete cytokinesis as observed in polytrophic meroistic species and lower hemipterans or by cellular fusion. We analysed the development of the ovary of *Dysdercus intermedius* throughout the five larval instars. Up to the third instar, non-dividing germ cells form a tight sphere in the ovariole anlage. In the center of the sphere phosphotyrosine containing proteins and f-actin accumulate and form the so-called PY-structure. With the molt to the fourth instar germ cells begin to divide asynchronously. Simultaneously, the PY-structure expands throughout the germarium. In the fifth instar the PY-structure retracts to an apical position, where it is maintained even in adult ovarioles. Germ cells that loose contact to the PY-structure cease proliferation. The basalmost of these cells enter meiosis to become oocytes and those located more apically endopolyploidise to function as nurse cells. Throughout larval development neither ring canals nor fusomes were observed. Taken together, these findings support the idea that the germ cell syncytium develops through cellular fusion.

Effect of *Toxoneuron nigriceps* polydnavirus gene expression in *Drosophila* germ-line

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Several parasitic wasps, when ovipositing in host insects inject proteins and a polydnavirus (PDV) that disrupt their immune reaction, development and physiology. Polydnavirus DNA is integrated as provirus in the parasitoid genome, excise and replicate in the calyx epithelium of the ovary to produce viral particles containing circular DNA molecules of different sizes. We focused on the *Toxoneuron nigriceps* (Viereck) (Hymenoptera, Braconidae), parasitoid of the larval moth *Heliothis virescens*, and its associated TnBV polydnavirus. We found that TnBV genome contains three genes (TnBVank1-3), coding for proteins with ankyrin repeats and showing 30% similarity with the *Drosophila* Cactus and other IkB proteins from different organisms. The viral proteins are small, 146 to 194 aa and, unlike other IkB proteins, do not contain the *N*-terminal conserved IKK target motif. We produced *Drosophila* transgenic lines carrying TnBVank1 and, using the Gal4/UAS system, we expressed the gene in the germ line and in somatic follicle cells. We observed that expression of the TnBVank1 viral gene in the germ-line induced a female sterility phenotype, while expression in follicle cells seemed to have no effect. Thus, with these experiments we could have disclosed a new function of Ikb-like proteins during *Drosophila* oogenesis, to be further characterised.

Functions of multiple sperm-stores in females of the damselfly *Ischnura senegalensis*

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Many insect females have multiple sperm-stores: i.e., bursa-copulatrix and spermatheca(e). The function of this multiplicity has been controversial. Arms' race theory suggests that multiple stores have evolved in females to avoid sperm competition. While cryptic female choice (sperm selection) theory suggests that multiple stores have evolved because of easiness for females to influence on paternity. We propose a new type of sperm selection mechanism within female sperm stores, which was suggested by sperm count conducted on the damselfly *Ischnura senegalensis*. Virgin females were allowed to copulate with a virgin male and then kept individually. Females were dissected on varying days after copulation. Bursa-copulatrix and spermatheca were taken out from the female and the number of sperm was counted under a microscope. We found that the number of sperm in bursa-copulatrix decreased with time, but the number of sperm in spermatheca did not, suggesting that the sperm mortality is higher in bursa-copulatrix. This result suggests that females discriminate against sperm in bursa-copulatrix, from which sperm may be displaced if she happens to recopulate later. Female sperm-storage organs may also function as spaces for the sperm discrimination (selection) as well as for the avoidance of sperm competition.

Regulation of juvenile hormone synthesis in mosquito: physiological, biochemical and molecular studies

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We aim to understand the regulation of juvenile hormone (JH) synthesis in mosquitoes, and how nutritional signals affect the activity of the neuroendocrine system. JH III synthesis is low immediately after adult emergence, increases in sugar-fed females and decreases after blood-feeding. Aedes-allatotropin makes CA in newly emerged females capable for JH biosynthesis. *Anopheles gambiae* PISCF-amide-allatostatin (allatostatin-C-type) is very effective inhibiting JH synthesis. By raising larvae under different nutritional diets two different adult phenotypes (large and small females) were generated. Teneral reserves (protein, lipids and glycogen) were significantly lower in small females. JH synthesis was significantly reduced in females emerged with low teneral reserves and stimulated by sugar feeding; only when nutrients are appropriate the CA becomes capable of synthesizing enough JH to activate reproductive maturation. Libraries were made from corpora allata-corpora cardiaca complexes from *Aedes aegypti* and *Anopheles albimanus.* More than 1850 clones have been sequenced. Enzymes involved in JH synthesis and other important regulatory molecules have been identified among these clones. Supported by NIH RO1 to FGN.

Phase response of the circannual rhythm in the varied carpet beetle, *Anthrenus verbasci*

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Anthrenus verbasci (L.) (Coleoptera: Dermestidae) shows a circannual rhythm in pupation. This rhythm continues under constant light-dark cycles and shows temperature compensation to some extent. Switching larvae to a different daylength accelerates or delays pupation, thus indicating that a change in photoperiod functions as a zeitgeber in this system. Phase response curves have been obtained for circadian rhythms in various organisms to characterize the oscillators involved in these rhythms. In the present study, we examined the phase response of the circannual rhythm in *A. verbasci*. Larvae were kept under short-day conditions (LD 12:12) at 20 ºC, and exposed to long-day conditions (LD 16:8) for 4 weeks at various phases. The exposure to long-day conditions produced a phase-delay or a phase-advance in the pupation rhythm, depending on the phase at which larvae were exposed to long-day conditions. Thus, a phase response curve for the circannual rhythm was obtained. The curve resembled that for the circadian eclosion rhythm in *Drosophila pseudoobscura*, and was classified into Winfree's Type 0. Therefore, there is a common feature in phase responses between the circadian oscillator and the oscillator involved in the circannual rhythm.

Cryptobiosis of the sleeping chironomid, *Polypedilum vanderplanki*: a unique mechanism to cope with an unpredictable drought

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Diapause is a period of arrested development in many insects to survive adverse environmental conditions. Astronomical information, mainly the change in daylength is employed to predict future environmental changes. The sleeping chironomid, *Polypedilum vanderplanki*, is a dweller in temporal pools in semi-arid regions in Africa. The small and shallow pools are easily ied up even no rain for a few days, and larvae in the drying pools are also desiccated completely but can revive after rehydration by the next rain. Such a developmental arrest with zero-metabolism is called "cryptobiosis" or "anhydrobiosis". It is of interest to know how *P. vanderplanki* larvae deal with an unpredictable drought. We found that trehalose plays an important role for successful induction and maintenance of cryptobiosis. We also demonstrated that rapid accumulation of trehalose coincided with loss of water from the larvae without cerebral regulation. This paper will summarize physiological and molecular mechanism inducing cryptobiosis in the sleeping chironomid, *P. vanderplanki.*

Molecular analysis of cross-talk between juvenile hormone and 20-hydroxyecdysone

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We discovered that the expression of juvenile hormone esterase gene from *Choristoneura fumiferana* (Cfjhe) is induced by JH I and the JH I induction is suppressed by 20-hydroxyecdysone (20E). Cloning and analysis of the promoter region of Cfjhe gene identified a 30 bp region that supports both JH and 20E response observed for the Cfjhe gene. This 30 bp sequence contains a direct repeat element with a 4-nucleotide spacer and is designated as JH response element (JHRE). A reporter gene placed under the control of JHRE is induced by JH in a dose- and time-dependent manner in CF-203, L57, HvE1s and Aa23 cells, and 20E in the presence of EcR was able to suppress JH induction. Nuclear proteins isolated from all four cell lines bound to JHRE. Protein kinase C-mediated phosphorylation decreased the binding of nuclear proteins to JHRE. JH or calf intestinal alkaline phosphatase-mediated dephosphorylation increased the binding of nuclear proteins to JHRE. The results from these studies suggest that the JH action and its cross-talk with 20E involve multiple signal transduction mechanisms.

Comparisons of neural development in insects and crustaceans

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Several lines of evidence have been used to homologize specific neurons and neural precursor cells between insect species and between insects and crustaceans. Our lab has recently characterized the expression pattern of Pax3/7 ("pairberry") family members between multiple arthropod species, and I will present data obtained from these studies that furthers our understanding of the evolutionary relationship of neural patterning and development in insects, and between insects and crustaceans. I will also present recent data on how the Notch/Delta signalling system, so widely used for neuroblast formation in insects, is also used for other early patterning functions.

The evolution of germline development and axial patterning

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While germline development and early axial patterning are well understood in the model insect, *Drosophila melanogaster*, it is clear that many interesting evolutionary differences between species exist in these processes. I will review my lab's progress in understanding the nature of these evolutionary changes, how they might have occurred, and their consequences on subsequent development. I will focus on studies of axial patterning involving maternal factors in the grasshopper, Schistocerca americana, the amphipod, Parhyale hawaiensis, and the fruit fly, *Drosophila* melanogaster. I will also present data on the similarities and differences in the behaviour of germ cells between insects, and their sister taxa, the crustaceans.

Signal transduction in the corpora allata of adult *Heliothis virescens*

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The concerted action of juvenile hormone and allatotropin (AT) is necessary for egg development in *Heliothis virescens*, which is mediated by second messengers in the corpora allata (CA). The Ca^{2+} ionophore, A23187, stimulated JH production, as did *Manduca sexta* allatotropin (Mas-AT). Thapsigargin, which increases intracellular Ca^{2+}, significantly stimulated CA activity. The membrane-permeable Ca^{2+} chelator, BAPTA/AM antagonized effects of thapsigargin and Mas-AT, suggesting that Mas-AT likely affects CA activity by increasing intracellular Ca^{2+}. The inhibitor of IP_3-induced Ca^{2+} release from intracellular stores, 2-APB, had no effect on *H. virescens*. 2-APB also failed to affect Mas-AT activity, indicating that 2-APB may not act as an efficient Ca^{2+} release blocker. Modification of intracellular Ca^{2+} concentration clearly affects CA activity, however, CA are insensitive to changes in extracellular Ca^{2+}, suggesting efficient Ca^{2+} homeostasis. The diacylglycerol (DAG)-kinase inhibitor R59022 no effect on CA activity, but it potentiated effects of Mas-AT. This indicates that DAG might serve as a second messenger. PDBu, the phorbolester activator of protein kinase C (PKC), did not affect CA activity when applied alone or in combination with Mas-AT. In most animal there are several, tissue-specific, PKC isoforms present, which show different sensitivities towards inhibitory or stimulatory ugs, thus, requiring further experiments.

Juvenile hormone, broad, and adult development of the *Drosophila* abdomen

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In its developmental role, juvenile hormone (JH) acts to direct ecdysone action during the molt so that it does not cause the switch in programs that is necessary for metamorphosis. Broad is an ecdysone –induced transcription factor that is necessary for pupal development. Both its appearance and disappearance in polymorphic epidermis is prevented by JH, but not in imaginal discs. When misexpressed during either the larval or the adult molt of *Drosophila* at the beginning of new cuticle formation, Broad causes the production of a pupal cuticle by activating pupal cuticle genes and suppressing either larval or adult cuticle genes. Studies with the upstream promoter of an adult cuticle gene Acp65a show that Broad-Z1 can suppress expression by the 236 bp minimal promoter. The effect of mutations within the putative BR-Z1 binding site is under study. We have found several mutants with P-element insertions in the regulatory region of CG14709, a putative ABC transporter, that in response to JH show the prolonged expression of BR in the abdominal imaginal cells but form normal adult abdomens. The role of this gene in the JH pathway is presently under study. Supported by NIH.

Summary diapause of redlegged earth mite, *Halotydeus destructor* (Acari: Penthaleidae) in Australia

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In winter dominant rainfall regions of southern Australia, redlegged earth mite, an introduced pest from South Africa, feeds on plants during the cool moist winter months. The hot dry summer is spent as a diapausing egg inside the cadaver of the adult female mite. Diapause is broken by exposure to high temperature in summer and the eggs hatch the following autumn, triggered by combined moisture and cold temperature requirements. The induction of diapause in spring was measured over several years at different sites, by dissecting mites weekly to record the proportion of diapause eggs present. Induction of diapause was found to be triggered by day-length and climatic factors and was relatively constant between years at any one site, but varied across southern Australia by up to six weeks. This was modelled as a single date for any given location which predicted the onset of diapause. The predictions were tested at different locations by dissecting mites weekly at locations across Southern Australia to record proportion of diapause eggs. At 88% of the 75 sites, diapause occurred within seven days of the predicted date.

The role of spermatophores as mating plugs that cause unsuccessful matings (pseudocopulations) in *Archisepsis diversiformis* (Diptera: Sepsidae)

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Copulations of non-virgin female *Archisepsis diversiformis* reared in captivity were of two types: successful (average duration 28,9 min., spermatophore transferred and inserted into the spermathecal duct) and unsuccessful (average duration 1.3 min., no spermatophore transferred). Only about half of the matings were successful, and females only mated when their eggs were relatively immature. Successful matings occurred after egglaying and in absence of a spermatophore in the vagina from a previous mating. Pseudocopulations, in contrast, occurred before egglaying, with an old spermatophore present in the vagina. Three hypotheses are suggested: 1- Spermatophores may function as nourishment for the female, her eggs or the sperm, because they are usually degraded, rather than discarded intact. 2- Female retention of the spermatophore may function as a mating plug, preventing insemination when she is not ready to mate and saving energy she would otherwise have to spend trying to physically reject or discourage undesired males. 3- The male leaves the spermatophore as a mating plug to prevent future males from copulating successfully. It is not clear whether females are capable of discarding fresh, intact spermatophores.

Comparative aspects of dorsoventral axis formation in *Tribolium* **and** *Drosophila*

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The pupal RNAi technique has allowed a detailed functional comparison of maternal and zygotic dorsoventral patterning genes from *Drosophila* and *Tribolium*. As in *Drosophila* the dorsoventral axis of *Tribolium* embryos relies on a maternal signaling cascade, which requires a Toll-like receptor and a Dorsal-like NF-kB transcription factor. The loss-of-function of *Toll* or *dorsal* lead to apolar, completely dorsalized phenotypes, which are strikingly similar to the corresponding *Drosophila* mutant phenotypes. In contrast, the major zygotic patterning system responsible for the subdivision of the ectoderm in insects and vertebrates, the *decapentaplegic/short gastrulation* system (BMP/chordin in vertebrates) has a more fundamental role in *Tribolium* than in *Drosophila*. A knockdown of *short gastrulation* in *Tribolium* leads to strongly dorsalized embryos. In *Drosophila* the Toll-Dorsal system apparently has overtaken some functions which were fulfilled by the *decapentaplegic /short gastrulation* system in more ancestral insects. Thus, the relative importance of the two patterning systems has changed in evolution.

Follicle cell differentiation in bgcn mutant *Drosophila* ovaries

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Mutations in the *Drosophila benign gonial cell neoplasm (bgcn)* gene affect germ cell differentiation and proliferation in both sexes. For normal gametogenesis to occur, *bgcn* function was shown to be essential in the germ line. Loss of *bgcn* function arrests ovarian germ cells at an early developmental stage that combines characters of stem cells, cystoblasts and early cystocytes. The development of somatic follicle cells, however, proved to be independent from germ cell differentiation to a large extent. Follicle cells always adopted epithelial polarity. By using cell-type specific reporter gene - and enhancer trap - lines and various antibodies we were able to identify a number of correctly specified follicle cell types in *bgcn* mutant ovaries. Among them were polar follicle cells, stalk cells and centripetal follicle cells. Even more surprising, the majority of follicle cells in mutant ovaries underwent terminal differentiation, i.e. they stopped their mitotic activity, became polyploid and finally secreted a multilayered eggshell with ultrastructural properties comparable to those of wild type eggs. Thus, follicle cells in *bgcn* mutant ovaries reveal an intrinsic developmental program that appears to be largely uncorrelated to germ cell differentiation.

Genomic and non-genomic action of ecdysteroid: both are required for completion of programmed cell death in the anterior silk gland

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Programmed cell death (PCD) of the anterior silk glands (ASG) of the silkworm, *Bombyx mori*, is triggered by 20-hydroxyecdysone (20E) with exhibiting serial responses in order, i.e., blebbing, cell shrinkage, nuclear condensation, DNA fragmentation and nuclear fragmentation. The blebbing and cell shrinkage are under the control of genomic action of 20E. Among early genes, EcR B1, E75A, BHR3 and BR-C are up-regulated, while EcR-A and E75B are not. In addition to the genomic action, nuclear fragmentation and DNA fragmentation are controlled by a non-genomic action of 20E through its membrane receptor. The partially purified putative membrane receptor exhibits a high affinity to ponasterone A. A PKC inhibitor and a caspase 3 inhibitor equally suppressed the nuclear fragmentation as well as DNA fragmentation, and nether calcium ionophore nor phorbol ester mimicked the 20E action, indicating that the down-stream of the putative membrane receptor includes Ca^{2+}-independent protein kinase C and caspase 3. Combined results indicate that 20E acts through two distinctly different pathways, and the PCD is accomplished by an integration of the genomic and non-genomic actions.

Roles of pigment-dispersing factor immunoreactive neurons in circadian rhythms and photoperiodism in the blow fly, *Protophormia terraenovae*

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Adult females of the blow fly, *Protophormia terraenovae* show clear circadian activity rhythms and photoperiodism controlling reproductive diapause. The present study examined roles of pigment-dispersing factor-immunoreactive (PDF-ir) neurons in circadian activity rhythms and photoperiodism by electron microscopy and ablation experiments. Electron microscopy has revealed that at a region of the dorso-lateral protocerebrum PDF-ir fibers form output synapses with neurons in the pars lateralis (PL), which are necessary for diapause induction. PDF immunoreactivity was detected exclusively in large dense-core vesicles but not in synaptic vesicles. Ablation of a region at the anterior base of the medulla in the optic lobe abolished circadian rhythmicity under constant darkness in about 30 % of flies. There are 4 small and 4 large PDF-ir cells at the anterior base of the medulla. The more small PDF-ir cells were removed, the more flies showed arrhythmicity. The same operation invalidated photoperiodism. Diapause incidence was low both under long-day and short-day conditions irrespective of the number of PDF-ir cells remained. In the present study synaptic inputs from PDF-ir neurons to PL neurons were demonstrated. It is suggested that the small-type PDF-ir neurons are indispensable to circadian activity rhythms, and regions by PDF-ir cells play important roles in photoperiodism.

Involvement of *timeless* gene in photoperiodic induction of diapause in drosophilid fly, *Chymomyza costata*

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In drosophilid fly, *Chymomyza costata*, photoperiodic response and circadian rhythmicity seem to be controlled by common elements. Non-photoperiodic-diapause (npd) mutants which have lost photoperiodic response for diapause induction show arrhythmic behaviour. We cloned *period* and *timeless* genes from wild-type and npd strains. Abundance of mRNA of both clock genes in adult heads of wild-type was low in daylight and high at night with circadian variation. In npd mutants, transcription of *period* was arrhythmic and *timeless* was suppressed. Genetic linkage analysis demonstrated that *period* was not directly related with critical daylength in photoperiodism but deficient allele of *timeless* was strictly linked with loss of photoperiodic response. These results indicate that *timeless*, important oscillator of circadian clock, is involved in photoperiodic time measurement system. Larvae of this fly enter diapause when they grow under subcritical short daylengths. Cell-growth and differentiation for imaginal development are interrupted at 3rd-instar under short days. *timeless* possibly plays important function in photoperiodic control of cell proliferation.

Agonistic female responses to insemination: cryptic choice or self defence?

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Bed-bugs are unique amongst dioecious organisms in that they show traumatic insemination. When the male stabs the female with his intromittent organ sperm and seminal fluids are introduced into the female's haemocoel, but so are opportunistic bacteria. Several authors have suggested that the resultant immune response functions to select sperm in a manner that enhances female fitness. We present the results from experimental studies that examine the consequences of this mode of insemination and the function of the female's physiological response to it, given that microbes are also transmitted during insemination. Our results provide an insight into how selection via sexual conflict shapes the evolution of secondary sexual traits.

Egg start: from stem cell to cyst to oocyte

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Germline stem cells (GSCs) reside at the tip of the *Drosophila* ovariole in a niche that serves as a model for understanding how stem cells are regulated by BMP, Hedgehog and other signals. After GSC division, one daughter remains in contact with cap cells, a major niche component, while the other moves away and under the control of the *bag-of-marbles (bam*) gene divides synchronously to form a cyst of interconnected cells containing an asymmetric actin cytoskeleton (fusome). Forming cysts interact with somatic inner sheath cells, and later with the pre-follicle cell daughters of distally located somatic stem cells (SSCs). During the cyst stage we find that selected organelles and specific macromolecules are actively transported into the future oocyte to form the Balbiani body. We also find that a subpopulation of inner sheath cells is part of a previously undescribed stem cell lineage, and we name these cells "escort cells." Escort cells accompany germ cells as they leave the niche until they are replaced by pre-follicle cells. During much of this time, germ cell cysts retain the capacity to de-differentiate into GSCs if placed in an appropriate environment such as the larval gonad. We have studied the regulation of escort stem cells, their role in the growth and differentiation of germ cells, and their interactions with GSCs and SSCs. These studies have been facilitated by a "gene trap" screen, using P and PiggyBac elements, that fuses proteins encoded throughout the genome with GFP in living flies.

The transition from vitellogenesis to choriogenesis during silkmoth oogenesis.

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In the silkmoth, *Bombyx mori*, follicle development occurs asynchronously and follicles of unique developmental stages are arranged as single rows in the developing ovarioles. Because the developmental periods of vitellogenesis and choriogenesis are very extended, isolation of follicles at many distinct developmental stages during these periods is feasible. This feature makes silkmoth oogenesis an attractive system for studies on the long-term developmental program that governs the terminal differentiation of ovarian follicles. An overview of the regulatory factors that are involved in the regulation of the transition from vitellogenesis to choriogenesis in the silkmoth will be presented. This transition is signaled by the decline of active ecdysteroids in the circulation and the induction of a regulatory cascade initiated by the orphan nuclear receptor BmFTZ-F1. Current research is focused on the regulatory roles of two key transcription factors that have postulated to be involved in the control of initiation of choriogenesis, BmGATAb and the orphan nuclear receptor BmE75C. Through the use of the yeast two-hybrid system, proteins interacting with these factors have been isolated. Their possible role in the transduction cascades controlled by the two transcription factors under investigation will be presented.

Male coercion, cryptic female choice, or both? A case study from spotted cucumber beetles

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Debate continues over the degree to which males and females control the paternity of their offspring. Reproduction in *Diabrotica undecimpunctata howardi*, the spotted cucumber beetle (Coleoptera; Chrysomelidae), demonstrates how male coercion and female choice can operate simultaneously. Females control access to their bursas by constricting vaginal musculature. Males use copulatory courtship and cuticular odors to "coax" females into relaxing their vaginal muscles to permit complete penetration. When females do not cooperate, some males attempt to overpower females or exhaust them in order to force sperm through their vaginal tract. Sperm obtained in this way remains in the bursa and is not moved by the female into the spermatheca. But when an attractive male subsequently deposits a complete spermatophore into the bursa, sperm from the rejected male is transported to the spermatheca along with the sperm of the accepted male. Coercive tactics can be so successful that up to 56 % of the offspring are sired by rejected males.

Role of [His^7]-Corazonin in the control of phase polyphenism in locusts

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A neuropeptide, [His^7]-corazonin, present in the brain and corpora cardiaca of locusts is known to induce dark colors in locusts and grasshoppers. The present paper reviews the highlights of our locust project on the function of this neuropeptide in the control of phase-related characters. [His^7]-corazonin was first isolated from the American grasshopper *Schistocerca americana* without known function by J.A. Veenstra (1991), and recently we demonstrated that it induces black patterns in the nymphs of this species. Solitary adult locusts are characterized by large F/C (hind femur length / head width) and small E/F (forewing length / hind femur length) ratios compared with gregarious adults. Injection of [His^7]-corazonin into solitary (isolated-reared) nymphs caused a shift in these ratios towards the values typical for gregarious adults in the migratory locust *Locusta migratoria*. Gregarious adults are known to have fewer coeloconic sensilla (chemoreceptors) on the 8th antennal segment than solitary adults. Injection of [His^7]-corazonin was found to mimic the effect of crowding on development of this sensillum. Similar effects on morphometrics and antennal sensilla were also observed in the desert locust *S. gregaria*. These results suggest that [His^7]-corazonin plays an important role in the control of phase polyphenism in locusts.

Ecdysteroid titers and expression of ecdysteroid receptor during vitellogenesis in the soft tick, *Ornithodoros moubata* (Acari: Argasidae)

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A blood meal is essential for tick reproduction and results in the transmission of various pathogens. Therefore understanding the reproductive processes are essential for tick control. Our research indicates the involvement of two factors, one a peptide and the other likely a steroid, in the regulation of vitellogenesis. Here we present data indicating a role for ecdysteroids, as the second factor, in the regulation of vitellogenesis of *O. moubata.* Injections and topical treatment with high concentrations of ecdysteroids stimulated an increase in vitellogenin and egg laying. Vg synthesis occurs in both engorged mated and virgin females, but is not activated in virgin females sufficiently for eggs to mature. Engorged mated females show three ecdysteroid peaks (previtellogenic, oocyte maturational, and preovipositional phases) in the hemolymph and four peaks (previtellogenic, oocyte maturational, preovipositional and ovipositional phases) in the whole body extracts. Virgin females have no ecdysteroid peaks and do not lay eggs. The ecdysteroid receptor of *O. moubata* has been identified and receptor expression appears to coincide with vitellogenin up-regulation, egg maturation and oviposition. These results suggest ecdysteroids induce vitellogenin synthesis, egg maturation and oviposition in *O. moubata*.

Generation of cell diversity in the developing CNS of *Drosophila*

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Development of the central nervous system (CNS) involves the transformation of a two-dimensional sheath of uniform ectodermal cells (the neuroectoderm) into a three-dimensional structure in which particular numbers of various neuronal and glial cell types become arranged in a reproducible pattern and establish specific connections. *Drosophila* is a suitable model organism to study the mechanisms that lead to pattern and cell diversity in the CNS. One of the first steps during neurogenesis in the *Drosophila* embryo is the delamination from the neuroectoderm of a stereotype population of neural stem cells, called neuroblasts (NBs). Each NB acquires a unique identity, which is reflected by the time and position of its delamination from the neuroectoderm, the combination of genes it expresses, and by the production of a specific cell lineage. Specification of a unique NB fate largely depends on positional information in the neuroectoderm. In the developing ventral nerve cord NBs arising from corresponding segmental positions generally give rise to very similar lineages. However, some of these serially homologous NB lineages express distinct segment-specific characteristics. Segmental diversification of NBs is much more pronounced in the developing brain. Aspects of the specification and development of NB-lineages in the embryonic ventral nerve cord and processes of early patterning in the brain will be presented.

Photoperiodic modulation of circadian rhythms in crickets

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"The mechanism of photoperiodic time-measurement remains to be clarified, although hypothetical models have been proposed. Most of the models that incorporate circadian clocks assume that parameters of the clock are rather stable irrespective of given photoperiods. We analyzed photoperiodic modulation of circadian locomotor rhythms in crickets, *Teleogryllus emma* and *Modicogryllus siamensis*, of which post-embryonic development is known to be regulated by photoperiods. Short-days accelerate it and long-days decelerate in *T. emma*, although reversed effects are observed in *M. siamensis*. The circadian waveform of their locomotor rhythms was highly dependent on the photoperiod during the post-embryonic development. In *T. emma*, the length of subjective night was longer when exposed to longer nights. In *M. siamensis*, adults always showed nocturnal rhythms, while nymphs were diurnal in long-day conditions but nocturnal in short-day conditions. Results of experiments using *Gryllus bimaculatus* suggest that the photoperiodic modulation occurs at the circadian clock and coupling of the clock is important for the maintenance of the modulated waveform. The possible involvement of the photoperiodic modulation of circadian system in the photoperiodic time-measurement mechanism will be discussed.
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Towards functional analysis of oogenesis in *Tribolium castaneum*

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Tribolium castaneum has ovaries of the telotrophic meroistic type, i.e. the nurse cells are not included into the follicle, but remain in the anteriormost compartment of the ovariole, the tropharium. In contrast to *Drosophila*, germline cells are mitotically inactive in adult *Tribolium* beetles - cluster formation takes place during larval and pupal stages only. To date, little is known about the mechanisms underlying the differentiation process of germ cells into oocytes and nurse cells in this species. By morphological criteria, several pro-oocytes, located posteriorly in the tropharium, can be clearly distinguished from nurse cells. The pro-oocytes enter the vitellarium one by one and differentiate into oocytes. Every oocyte remains connected to the nurse cells via the so-called nutritive cord, a long protrusion formed by the anterior pole of the oocyte. Using RNA interference, we have initiated loss-of-function studies on *Tribolium* oogenesis. Females injected with *gbb* (*glass bottom boat*, a Bmp-orthologue) double-stranded RNA are completely sterile, and oocyte development is arrested at a premature stage. Analysis of injected females indicate that *gbb* is also involved in the organogenesis of the ovary.

Use of in vivo imaging of neurons to study hormone action on the insect CNS

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During insect metamorphosis most larval tissues degenerate and adult organs are formed from the diploid cells sequestered in the imaginal discs and imaginal nests. The CNS is more complicated than other tissues, however, because many of the larval neurons persist through metamorphosis and undergo cellular remodeling, allowing them to perform new functions in the adult. Ecdysone and juvenile hormone are the two metamorphic regulators of cellular remodeling. They regulate the expression of intrinsic programs for pruning and regrowth of denitic and axonal arbors in a given neuron but they also are responsible for extrinsic cues produced in the cell's immediate environment which interact with the cell's intrinsic programs. Our goal has been to determine the intrinsic and extrinsic factors that regulate cellular remodeling. The talk will focus on the use of in vivo imaging of remodeling neurons as a tool for examining how ecdysone and juvenile hormone regulate this process. Mutants, RNA interference and dominant negative receptors have been used to disrupt hormone response in either individual neurons or their cellular environment. These experiments indicate that remodeling involves a complex choreography of interactions between a neuron and the cells that it contacts.

Neuronal lineages and the development and evolution of the insect CNS

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The neurons in the segmental ganglia of insects arise from a stereotyped number of stem cells called the neuroblasts (NBs). In basal insect groups the NBs produce all of their progeny neurons during embryogenesis, but in insects that have complete metamorphosis many of the NBs split their neurogenesis into embryonic and postembryonic phases, to generate neurons for the larval and adult nervous systems, respectively. Although the early born neurons coming from each NB have been identified, the bulk of the neurons produced by each stem cell remain unknown. With this in mind, we undertook an extensive clonal analysis in *Drosophila* in which we determined the initial projection patterns for the late born neurons in all of the thoracic lineages. The late-born cells are remarkably homogeneous in their projection patterns and suggest functional restrictions amongst the lineages. For example some neuroblasts make interneurons that project only to presumptive leg neuropils whereas others project to presumptive flight neuropils. Since the arrays of NBs have stayed remarkably stable through the evolution of the insects, this description may provide the foundation for a better understanding of the evolution and diversification of the insect CNS.

Regulation of matrix metalloproteinases during metamorphosis of the lepidopteran *Galleria mellonella*

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Reorganization of the extra cellular matrix during tissue differentiation and metamorphosis is predominantly mediated by matrix metalloproteinases (MMP´s) which are regulated by tissue inhibitors of metalloproteinases (TIMP´s). More than 27 MMP and 4 TIMP´s homologues member of these families have been reported from vertebrates[1]. Recently, the first MMP and TIMP homologues have been reported from *Drosophila* [2,3] . Here we report our recent progress in identification of MMP´s in the lepidopteran *Galleria mellonella* and we provide evidence for the regulation of this endogenous metalloproteinases by a novel insect metalloproteinases inhibitor (IMPI) which represents the first non-TIMP-like metalloproteinaseinhibitor identified in animals[4]. Cloning and expression of the IMPI as well as analysis of the signalling pathway mediating its induction led to the discovery of a feedback loop regulation mechanism in *G. mellonella* that controls endogenous MMP´s mediating remodelling of the extra cellular matrix during metamorphosis.

Molecular cloning and recombinant expression of antimicrobial peptides from house fly, *Musca domestica*

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Two antimicrobial peptide genes, the cecropin and defensin, were cloned by from the houseflies, *Musca domestics.* The full length of the cecropin cDNA was 403 bp and the open reading frame of the cDNA encoded a 63 amino acid procecropin. A potential signal sequence of twenty-three amino acids was identified, ending with the residues QSEA. A single cleavage reaction at Ala-23 therefore would generate the mature forty-amino acid *Md*-Cec peptide. The N-terminal portion of the molecule is highly basic, while the C-terminal domain is predominantly hydrophobic. The sequence identity with other cecropin was between 80% and 97%. The open reading frame of the defensin encoded a 92-amino acid peptide, which contained an NH_2-terminal signal sequence (1~22) followed by a propeptide and the mature peptide (53~92). The sequence identity with other insect defensin is between 51% and 73%. The mature peptide, with a predicted molecular weight of 4.0 kDa, and pI of 8.69, has 1 negative charged amino acid and 4 positive ones. The cecropin and defensin were expressed in *E. coli* and yeast and the recombinant peptides in yeast had the activity against several Gram negative and positive bacteria. The recombinant cecropin and defensin were confirmed by amino acid sequencing.

Bridging the gap between molecular and cellular functions of axon guidance molecules

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Over the last decade, many molecules that play key roles in axon guidance have been identified. However there remains a substantial gap between our understanding of the biochemical functions of such molecules and how their effects are translated into specific guidance decisions. To tackle this problem, we have chosen the sensory nervous system of the *Drosophila* embryo as our model. The identities of the growing axons, their cellular guidance cues and key axon choice points are known in detail in this system and *Drosophila* offers an arsenal of powerful molecular/genetic tools. Using the candidate gene approach, we have identified roles for a number of known axon guidance molecules in sensory axon guidance. These include Slit and its receptors, the Robo family, Semaphorins and their receptors, the Plexins and the cell adhesion molecule Neuroglian. Our results have shed new light on the cellular mechanisms by which these proteins mediate axon guidance. We now seek to determine how the growth cones of the sensory neurons integrate these multiple guidance signals at particular axon choice points.

Molecular cloning and expression patterns of the molt-regulating transcription factor, Hhr3 from *Helicoverpa armigera*

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A 1668 bp cDNA encoding a molt-regulating transcription factor, hormone receptor 3 (HHR3) was cloned from *H. armigera,* which deduced a protein with 556 amino acids. The isoelectric point of this protein was pI 6.52 and the molecular mass was 62 kDa. A DNA-binding region signature of nuclear hormone receptor was found from No. 107 to 133 amino acids. A possible transmembrane helice from outside to inside was found from No. 72 to 90 amino acids. Five bands of HHR3 were detected from the larvae by northern blotting. Among of them, band 1 and 2 were the dominant populations. The expression patterns of HHR3 *in vivo* were variable upon developmental stages and tissues. Results suggested that bands of 1 4 of HHR3 was only shortly expressed during molting which suggested that they were involved in the regulation of molting cascade, whereas, band 0 was observed in both molting and feeding larvae. Band 1 and 2 of HHR3 could be inducted from epidermis by RH-2485. The cDNA of HHR3 was expressed in *E. col.* A reconstructed HHR3-NPV was prepared and the efficacy to pest insect was studied. The work was supported by grants: 3008002, 30330070 and 2001AA214031-1-2.

A possible pathway for diapause termination in *Helicoverpa armigera*

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Diapause hormone (DH) is a 24 amino acid peptide amide which induces embryonic diapause in *Bombyx mori*. A DH-like neuropeptide, which is 62.5% identical with *B. mori* DH was found in the PBAN cDNA from the suboesophageal ganglion (SG) of the bollworm, *Helicoverpa armigera* (Har). Using quantitative RT-PCR and ELISA, we observed that expression of the Har-DH-PBAN mRNA in the SG and the DH-like immunoreactivity in hemolymph of nondiapause-type pupae was significantly higher than in diapause-type pupae. Injection of the amidated Har-DH into diapausing pupae effectively promoted development at all stages of diapause. Immunocytochemical results indicated that the level of DH gene expression and axonal release are related to the diapause decision. Ecdysteroidogenesis in prothoracic glands (PGs) was stimulated by synthetic DH *in vivo* and *in vitro*, and labeled DH bound to a receptor on the PG, thus suggesting that DH breaks diapause by activating the PG to synthesize ecdysone. Furthermore, the response of DH in terminating diapause was temperature-dependent. Decerebration experiments showed that the brain can control pupal development through the regulation of DH, and DH can terminate diapause and promote development without the brain. This result suggests a possible mechanism of response for the DH signal in *H. armigera*.

Effects of circadian phases on sperm competition in *Drosophila melanogaster*

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Females in almost all animal groups copulate with multiple males. In addition, female insects have spermatheca to store sperm. Hence, sperm competition is an important component of fitness in males. Many insects show daily rhythms in their mating. The mating activity of *Drosophila* is controlled not only by an endogenous clock, but also by light itself. It has been suggested that circadian clock is important for sperm release in *Drosophila melanogaster*. In this study, sperm precedence was studied by employing various mutant strains and a wild type strain. These strains were entrained in two different LD conditions (anti-phased for 10hrs). The preliminary data showed the circadian phase did not affect the outcomes of sperm competition. The underlying mechanisms were also investigated.

Gene expression during diapause initiation of the Colorado potato beetle, *Leptinotarsa decemlineata*

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Duration of diapause can be influenced by environmental factors prior to the initiation of true diapause. To clarify the molecular mechanism(s) regulating diapause initiation and how the duration of diapause is determined, early diapausing beetles are being screened for diapause regulated genes. To date, six genes have been isolated and their expression investigated in prediapause and early diapausing beetles. Diapause-associated transcripts 1 & 2 (DAT-1 & 2) are expressed beginning on day 3 after adult emergence in short day adults. DAT-3, HSP70A and a small heat shock protein are expressed at the initiation of diapause. An unidentified nondiapause upregulated transcript is expressed in day 1 nondiapausing adult female beetles. Genomic structures of several of these genes have been determined in order to identify possible promoter sites and to identify possible noncoding RNAs within the introns. Six possible promoter sites have been identified within 700 bp upstream of the TATA box of DAT-3. A possible ncRNA associated with these diapause regulated genes has also been isolated, indicating another layer of complexity in the regulation of diapause.

Development, evolution and function of a highly conserved insect stretch receptor

DJ Merritt, J Rice, R Herbert, S Shrader

Paper not available at time of production

Extremely female-biased sex ratios in a parasitoid wasp, *Melittobia australica* (Eulophidae), in relation to emergence pattern and lethal male combat

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The parasitoid wasp *Melittobia australica* mates on their hosts. Sex ratio is predicted to follow local mate competition (LMC) theory. Notwithstanding that LMC models predict the sex ratio will increase from female-biased toward a 1:1 ratio with increasing numbers of foundresses, the observed female-biased sex ratios (about 2% males) hardly increased in response to an increased number of foundresses. In this paper, we report sex ratios of two foundresses who sequentially parasitized the same host. We distinguished them individually using microsatellite DNA marker. The offspring sex ratios of both were extremely female-biased (first foundress, 1.1%; second foundress, 2.7%). Emergence period of offspring produced by the two foundresses were widely overlapped each other and their sons intermittently emerged during a prolonged period at constantly low late. In addition, an arena combat experiment showed later emerging males were killed by former ones even when they were brothers. These results suggest that each foundress should produce small number of male eggs at some intervals in order to prevent from producing wasting males that were to be killed by lethal male-male combat. This combat may be one of factors that explain the extremely female-biased sex ratio even with increasing foundresses in this species.

Reversal of the left/right-asymmetry in male genitala: a non-rare modification in Asian sericine chafers (Coleoptera: Scarabaeidae: Sericini)

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The examination of genital morphology is widely accepted as a principal diagnostic feature for species' identification in most insect groups. During an extensive amount of examinations of aedeagi among sericine beetles (Coleoptera: Scarabaeidae) it was noticed that the reversal of left/right-asymmetry in male ectodermal genitalia, a phenomenon rarely reported from insects in literature, seems to cumulate within this group. Among several hundreds of species with tens of thousands of specimens examined, so far from Asia only, seven species showed a left/right asymmetry reversal. The modified structures were parameres and phallobasis, only, showing the mirror imagine from the original state. The portion of reversal reported range from 0.003 to 0.5 percent of the studied male individuals. Since in the modified specimens, neither the asymmetric mandibles nor the asymmetric sutural junction of elytra were affected by any modification of normal left/right asymmetry, it should be supposed that the reversal is gained during abdominal segmentation.

Self regulation of the sun collector efficiency of *Archeoprepona menander* (Charaxinae – Lepidoptera)

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In butterflies favouring direct absorption of solar energy through their wings, The absorption by the black areas situated at the wing bases provide the body with the most part of the exogenous energy. Absorption is very high over the whole solar spectrum (a ~ 0.98). This absorption ids due to both the dendritic structure of the scales and the melanin pigments. In the infrared range, the absorptivity reveals two strong peaks at 3µm and 6µm, then remains more or less constant at a relatively high value (~0.4) up to 50 µm. This situation contains the outline for a remarkable self regulation phenomenon of the radiative balance, allowing the stabilization of temperature in the survival area of the butterfly. The first absorption peak, centered on 3 mm, i.e. between the solar radiance and the thermal emission spectra, is not involved in the radiative balance at these temperatures. However, the second one on the edge of the emission spectrum, plays a key role as a thermal regulator. Its marginal situation is such that according to the wing temperature, it overlaps or not the emission spectrum, modifying the collector efficiency, and thus its temperature.

Characterisation of a Type IV secretion system identified from the insect endosymbiont *Wolbachia pipientis.*

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Wolbachia is a reproductive parasite of as many as 70% of all known insect species, and imposes a range of effects upon the host including parthenogenesis, cytoplasmic incompatibility and selective male killing. Little is known about the molecular events that impose these effects. Using the recently completed genome sequence of *Wolbachia pipientis*, 12 ORFs were identified, on the basis of sequence homology and synteny, as putative Type IV secretion system genes. Type IV secretion systems are known to facilitate the export of bacterial effector molecules, protein or DNA, into a host environment to establish a favourable environment in which the bacterium may reside. Such systems are utilised by plant biotechnologists for the genetic transformation of plants using *Agrobacterium* and are responsible for the secretion of toxins from a range of medically important pathogens. We are interested to know if the putative *Wolbachia* Type IV secretion system is responsible for delivering effector molecules into host insect environment and thereby inducing the observed host effects. We provide here evidence that these ORFs are expressed in *Wolbachia* and discuss what *Wolbachia* proteins interact with components of the putative secretion system identified from our initial yeast-two hybrid experiments.

Number of sexual alleles in two Amazonian *Melipona* bee population

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Sex determination in stingless bees (Hymenoptera, Meliponini) involves the multi-allelic gene *xo* or *csd* (Complementary Sex Determine) locus. Diploid females are heterozygous for the sex locus, whereas males are haploid and arise from unfertilized eggs. To count and monitor the number of *xo* alleles is a practical method to detect inbreeding and the consequence of arising diploid males, in a limited population. This work was carried out on a meliponary located in Manacapuru, State of Amazonas, Brazil, were we monitored 33 *Melipona seminigra* and 202 *Melipona compressipes* colonies in a period of two years. In order to verify the female and male segregation we follow the methodology used by Carvalho (2001) obtaining the first brood cells disc from a young fertilized queen. The results show 16 alleles in both species indicating that the introduction of new fisogastric queens and the proximity to the native nests located in the near forest fragments serves to keep the sexual alleles variability and guarantees the colonies survival and artificial multiplications, and according to the proposle by Carvalho *et al.* (1995), it does help *Melipona* beekeepers maintain the variability of their colonies.

The evolution of new sex chromosomes of *Drosophila albomicans*

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The neo-X(3), neo-Y(3) arms of *Drosophila albomicans* and the 3rd autosome of *D. nasuta* are homologous. The former two are newly formed sex chromosome arms through fusion during speciation of *D. albomicans* about 500,000 years ago, and the last one remains to be an autosome in its sibling species *D. nasuta*. For the lack of recombination in male *Drosophila*, the neo-Y(3) is expected to accumulate deleterious mutations and degenerate. On the contrary, the neo-X(3) suffers higher selection force when presents in a male. Comparing homologous sequences from these chromosomes can be informative. Based on some pilot studies I did in the previous year, I've found the best way to approach this aim. A cDNA library of *D. nasuta* and the male genomic library of *D. albomicans* will be constructed. About 50 cDNA clones will be mapped by *in situ* hybridization on the salivary gland polytene chromosomes. A few important clones located on this particular part of the genome will be chosen. The corresponding genomic clones will be sequenced, and proper primers will be designed accordingly. By properly designed mating procedure, flies with homologous chromosome can be generated, and used for PCR-sequencing. Then, homologous sequences from the three groups of chromosomes will be obtained for comparison. Theoretical hypotheses about evolution rate, polymorphism, *etc.* will be proposed and tested.

Rapid cold hardening tactics in Antarctic arthropods

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Collembola and Acari form the large majority of Antarctic terrestrial arthropods. They are predominantly freeze avoiding and survive low winter temperatures by supercooling. In the summer, animals which are active and feeding run the risk of low levels of cold tolerance. Seasonally, most species are known to stop feeding and cold-harden prior to winter. However, a particular problem exists in summer habitats which are chronically close to 0ºC, as surface temperatures can approach the freezing boundary rapidly, leaving feeding animals vulnerable to freezing, while not providing sufficient cue to allow the activation of seasonal hardening tactics. Species such as the common Antarctic springtail, *Cryptopygus antarcticus*, have developed the ability to rapidly enhance cold-hardening over a period of hours, leading to a reduction in freezing point from c. -5 to below -25°C. This strategy allows individuals to take maximum advantage of any short warm spells for feeding and activity, thereby contributing to the remarkable abundance seen in some Antarctic terrestrial habitats, and the speed of response of the species to local climate change. We present data quantifying the rate of the cold hardening response and the environmental factors which initiate it, and provide an initial comparison with the tactics used by other polar species from habitats with different thermal characteristics.

Diversity and chromosome distribution of retroposons in *Anopheles gambiae*

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Mosquitoes are the first species group to demonstrate high diversity of retroposons. A detailed analysis of retroposons in the *Anopheles gambiae* genome identified 9 distinct clades. CR1 was the most diverse clade with 36 distinct families. In contrast RTE contained only two, but very large, families. Further analysis examined chromosomal distribution of the different types of elements to determine if insertion sites were a factor in determining inter-family or inter-clade differences. For example, the rDNA specific insertions observed with R1 elements, or co-amplification with tandem arrays of repetitive DNA observed with Juan elements. The distribution of all retroposons was shown to be non-random, but no specific inter-family or inter-clade trends were observed. High densities of retroposons were found in the sub-telomeric and gene poor regions of the chromosomes. Detailed analysis of retroposon dense and poor regions is in progress. Also, the retroposon distribution is being compared to other repetitive DNA distributions. For example, chromosome maps of all microsatellites have been constructed using “MSFind”, a program written to identify the start and end positions of all microsatellite repeats >10 bp.

Retroposon diversity and chromosome distribution in insects

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Aspects of polymorphism in the grasshopper *Orphulella punctata* (DeGeer) in Jamaica

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Polymorphism describes a population with two or more forms of a species existing at the same time, yet the rarest occurs at a greater frequency than that which can be maintained only by recurrent mutation. The geneticist, E. B. Ford, was among the first to describe polymorphism in the 1940s. Colour pattern polymorphism is common in acridid grasshoppers and has been studied in several temperate species. In the Jamaican grasshopper, *Orphulella punctata* (DeGeer), it is seen as a green-brown variation overlaid with patterns. A detailed classification of this polymorphic system revealed three colour morphs and ten patterns occurring in various combinations. Sampling in different rainfall zones showed a significantly higher frequency of green forms in wetter areas. Green and green-brown females are also significantly larger than brown females. Colour therefore, either plays a role in fitness, or has coevolved with size. Breeding experiments to analyse the genome structure, especially gene linkages, are in progress. Analysis of randomly polymorphic DNA is also being done to discover the relatedness of these forms, and give insight into their evolution. The aim is to understand the main factors keeping polymorphism stable within the disturbed habitats in which this species live.

Wing shape differences between migrant and resident monarch butterflies, *Danaus plexippus* (L), in Cuba

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The majority of migrant monarchs from eastern United States and southeast Canada migrate to Mexico, however, some of them migrate to Cuba and possibly to other areas of the Caribbean. Cuban migrants hatch in the southwest of Canada and eastern United States, and then engage in a southern trip of 4000 km to this Caribbean island. In Cuba, these migrants encounter the resident monarchs, which do not migrate and instead move between plant patches looking for nectar, mating partners, and host plants. Resident monarchs fly short distances and close to the vegetation, in contrast, migrant monarchs fly very long distances at high elevations. I explored whether flight behavioral differences between migrant and resident monarch in Cuba are associated with differences wing shape. To test if migrant and resident monarchs differ in wing shape, monarchs were collected for four years in Cuba. Fourier analyses was used to quantify wing shape differences between these two populations, as well as wing angular measurements. Results show that migrant and resident females differ in their wing shape. Female migrant monarchs have more elongated wings to reduce turbulence and drag during their long migration south and to enhance gliding. Cuban migrant monarchs are not an homogenous group in terms of their wing shape, either.

Sexual dimorphism and sexual selection in a highly dimorphic orb-weaving spider

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Spiders (Araneae) are the only free-living terrestrial taxon where extreme female-biased sexual size dimorphism is common. Current hypotheses for the adaptive significance small size ("dwarfism") in male spiders predict a small male advantage during mate search or during mating with cannibalistic females, or assume that sexual selection for large size due to interference competition is weak. Here, we examine selection on adult males of the highly dimorphic orb-weaving spider *Argiope aurantia*, using a multivariate approach to distinguish selection targeting different body components. Our results show that selection tends to favour longer legs during mate search. This is consistent with the pattern of sexual dimorphism in shape, and also results in net selection for larger males. Large male size is also favoured during interference competition over access to moulting females, the most common mating situation for males. Males that mate with mature, post-moult females face an 80% chance of a cannibalistic attack, but this does not result in selection on male body size. Thus, we find no support for any of the current small-male advantage hypotheses. In spite of extreme female-biased size dimorphism, net selection on adult male *A. aurantia* tends to favour larger rather than smaller males.

Life cycle synchronization of *Chrysolina aurichalcea* (Mannerheim) (Coleoptera: Chrysomelidae) to host phenology of *Artemisia princes*, with special reference to temperature and photoperiodic responses of egg diapause.

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C. aurichalcea has a univoltine life cycle with commonly two diapause periods of egg and adult. It is a very common insect and is widely distributed in the Japanese islands. Eggs hatch early spring and adults emerge in early summer. Adults enter summer diapause and the breeding season begins early autumn. Eggs laid in autumn enter winter diapause. Egg hatching is synchronized to the sprout emergence of host plant, *Artemisia princes*. The emergence begins at rather cold temperature near to 0-5**°C** prior to those of other weeds. The phenology of host plant varies drastically among geographic populations, i.e. evergreen in warmer area but deciduous in winter at cooler area. We examined the effects of temperature and photoperiod on the egg period. The intensity of egg diapause collected at same date among different altitudes increased with the decrease of winter temperature. The diapause intensity of the egg from transient zone was influenced by both temperature and photoperiod of not only eggs period but also their adults. Furthermore the effect seemed to vary with the different of their maternal age. The significance of such variations to seasonal adaptations under different climatic conditions and different host plant phonologies will be discussed.

Egg guarding and second oviposition as a plastic response to egg predation in a crab spider

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Females of a crab spider, *Lysiteles coronatus* guard their egg sac against other arthropods without taking food in folded leaves. This study determined the evolutionary factors of egg guarding and second oviposition by *L. coronatus* females. Egg survival was significantly lower when the mother was experimentally removed than when the mother was allowed to continue egg guarding. Predation by other arthropods is attributed to the major source of egg mortality that has led to the evolution of maternal egg guarding. Females lost about 30.2% of their weight for about a 40-day period of egg sac guarding, and they had degenerated ovaries at the termination of egg care. Such high cost of egg care would have prevented *L. coronatus* females from subsequent oviposition. However, when females were separated from their eggs and given sufficient food, they produced a second egg mass. The ability of second oviposition peaked at the midpoint of egg-guarding period and then reduced. In the field, a few females produced the second egg mass one month later than did ordinary females. Evidence available suggests that the potential of second oviposition is maintained as an adaptive plastic response to the loss of the first brood due to predation.

A toxic tale: evolution of the vertebrate toxin gene, a-latrotoxin, in black widow spiders

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The venom of black widow spiders (Theridiidae: *Latrodectus*) contains multiple neurotoxic proteins, including the vertebrate neurotoxin alpha-latrotoxin (a-LTX). Though the function, structure and gene sequence of a-LTX has been thoroughly characterized in one species of the genus (*L. tredecimguttatus*), the phylogenetic distribution and evolutionary origin of the a-LTX gene is unknown. In addition to a-LTX, venom of *L. tredecimguttatus* contains insect and crustacean-specific neurotoxic proteins possessing structural motifs and sequence homology at the amino acid and nucleotide level similar to a-LTX. We hypothesize that these different toxins (expressed by a single individual) are products of a gene family whose members arose via duplication events. Here we investigated the evolution of the vertebrate specific a-LTX toxin gene by sequencing a 700 bp fragment of this gene from six *Latrodectus* species in addition to constructing an independent species phylogeny for the genus using the mitochondrial gene COI. Our data indicates that a-LTX occurs across a broad phylogenetic spectrum of *Latrodectus* species, suggesting that this gene existed in the common ancestor of the genus and that it likely occurs in closely related genera. Across the sequenced region, we observed substantial nucleotide variation, translating into amino acid differences, suggesting the gene is rapidly evolving.

Seasonal changes in glycerol content and enzyme activities in the overwintering larvae of *Ostrinia furnacalis* located in northern and southern part in Japan

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Enzymes associated with glycogen metabolism and glycerol synthesis in larvae of *Ostrinia furnacalis* located in Shonai and Hyogo district, which are northern and southern part in Japan, respectively. The profiles of cold hardiness of natural populations of larvae of the northern and southern part of Japan, seem different. In the present study, seasonal changes in the level of glycerol, cold hardiness and enzyme activity in the overwintering larvae were investigated over the winter in 2002. Glycerol content and cold hardiness (survival rate at various treatment days under -15 °C) of the Shonai larvae were low in October, rapidly increased in November and December, peaked in January, while in Hyogo larvae, glycerol content and cold hardiness peaked in February. Glycerol accumulation was accomplished by activation of α-glycerophosphate dehydrogenase and polyol dehydrogenase with glyceraldehyde activity and inhibition of glyceraldehyde kinase in Shonai larvae.

Evolution of reproductive strategies of flour beetles (Coleoptera: Tenebrionidae)

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Although, they first evolved as saprophylic insects and naturally occur under the bark of trees, in dead wood and even in the nest of some Hymenoptera, *Tribolium* flour beetles have been adapted to stored products environment since several thousands years. In this study we aimed to observe if as a secondary adaptation to this ecosystem similar reproductive strategies were adopted by these species. Intra- and inter-specific allometric relationships were examined between several reproductive traits, e.g. egg mass, adult mass, development time and fecundity, in eight *Tribolium* flour beetles. Experiments were conducted under the same conditions for every species. Univariate and multivariate statistical analysis were first performed. Common life history traits were not observed between the different species and univariate analysis highlighted strong differences between the species for most traits examined. Some species presenting reproductive traits allowing an efficient development in the environment of stored products are likely to have occur in this ecosystem for longer periods than the others. A phylogeny of these species was established on DNA sequences of mitochondrial 16S rRNA gene. With this informative framework, the evolutionary pattern of these traits was studied using an explicit model of evolution and interspecific allometric relationships were examined, after controlling for phylogenetic independence.

Mechanisms of rapid cold hardening in *Drosophila melanogaster* - Are rapid changes in membrane phospholipids the explanation?

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Low temperature mortality is not always associated with freezing in insects. In many insects it may also occur at temperatures above the freezing point of the hemolymph. Reasons for this chilling injury could be irreversible phase transitions of membranes or protein denaturation at low temperature. Rapid cold hardening (RCH) is a phenomenon in which insects achieve increased tolerance of low temperature during a brief (minutes to hours) exposure to low, but sublethal temperatures. So far, RCH has been shown in several insect species including *Drosophila melanogaster* but no convincing physiological explanation of the RCH mechanism has been shown. In a series of experiments we have studied the membrane phospholipid composition in *D. melanogaster* when exposed to a cooling regime increasing the chill tolerance by RCH. Flies cooled from 25°C to 5°C at 0.1°C h^{-1} displayed a shift in the C16 and C18 fatty acids towards a higher degree of unsaturation. The results indicated that these changes were probably not caused by new synthesis of membrane phospholipids but rather on modifications of existing fatty acids. The same material was analysed for Hsp70 levels, but RCH induced no increase in this stress response. In conclusion, even though the observed shifts in membrane phospholipid unsaturation degree were small, they are likely to have contributed to the rapid cold hardening in *D. melanogaster*.

Evolutionary trade-offs between reproduction and dispersal in populations at expanding range boundaries

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During recent climate warming, some species have expanded their ranges northwards to keep track of climate changes. Evolutionary changes in dispersal have been demonstrated in these expanding populations and here we show that increased dispersal is associated with reduced investment in reproduction in populations of the speckled wood butterfly, *Pararge aegeri*a. Evolutionary changes in flight versus reproduction will affect the pattern and rate of expansion at range boundaries in the future, and understanding these responses will be crucial for predicting the distribution of species in the future as climates continue to warm.

Bacterium in the *Cytophaga-Flavobacterium-Bacteroides* phylum causes cytoplasmic incompatibility in spider mites

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Cytoplasmic incompatibility caused by *Wolbachia*, is widely observed in insects and mites. *Wolbachia* are also causative agents of parthenogenesis, feminization and male-killing. Recently, bacteria in the phylum *Cytophaga-Flavobacterium-Bacteroides* (CFB) have been reported to cause reproductive alteration in their hosts: parthenogenesis in parasitoid wasps (Zchori-Fein et al., 2001), feminization in false spider mites (Weeks et al., 2001), and cytoplasmic incompatibility in parasitoid wasps (Hunter et al., 2003). This group of bacteria is also called CLO (*Cytophaga*-like organisms) and is recently named as *Candidatus* Cardinium hertigii (Zchori-Fein et al., 2004). Our study aims to clarify whether CFB bacteria cause cytoplasmic incompatibility in spider mites. 16S rDNA sequencing data disclosed that CFB bacteria infect the spider mite, *Eotetranychus suginamensis*, *Oligonychus ilicis*, *Amphitetranychus quercivorus* and *Tetranychus pueraricola*. The CFB bacteria in the spider mites form a monophyletic group with those in the parasitoid wasps and the false spider mites. Crossing experiments between CFB-infected and uninfected colonies left reproduction unaltered in *O. ilicis*, *A. querciborus* and *T. pueraricola,* whereas in *E. suginamensis*, eggs hatchability and percentage daughters in the crosses between uninfected females and infected males were significantly lower than those in the reciprocal crosses. Therefore, CFB bacteria in *E. suginamensis* were related to the induction of cytoplasmic incompatibility.

Distribution and function of ankyrin domain proteins in the insect endosymbiont *Wolbachia pipientis*.

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The endosymbiotic bacteria *Wolbachia pipientis* infects a wide range of arthropods and filarial nematodes inducing a variety of phenotypes including parthenogenesis, male killing, feminization and cytoplasmic incompatibility. The recent annotation and sequencing of the first *Wolbachia* genome is providing new insights into the molecular mechanisms used by this symbiont to infect and manipulate their eukaryotic hosts. One of the main features of *Wolbachia*'s genome is the presence of an unusually high number of proteins containing ankyrin repeats. These domains are typically involved in protein-protein interactions and they have never been found before in such numbers in any prokaryote genome. In this work we have determined the distribution and expression of the different ankyrin repeat genes found in the sequenced *Wolbachia w*Mel genome in various *Wolbachia* strains that induce different effects in their hosts. We found correlation between the distribution of some ankyrin domain proteins and the induction of cytoplasmic incompatibility. Some non-CI inducing strains showed a deletion of 22 kb surrounding one of the ankyrin domain genes. This deletion affects 13 ORFs and is probably the result of a transposition event. The possible role of ankyrin domain proteins and their interaction with host proteins is also discussed.

Cuticular lipids of the tsetse fly, *Glossina pallidipes*

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Cuticular lipids play a critical role in preventing desiccation and in chemical communication. This poster will present data indicating that cuticular lipid mass may not be directly correlated with rates of water loss in laboratory-reared and wild-caught tsetse flies. This is in agreement with previous research conducted by others using *Drosophila melanogaster*. We did find correlation with various protocols indicating a treatment effect. We will present the identification of the major cuticular lipids of which hydrocarbons make up a relatively small amount. Hydrocarbons are thus probably more important in chemical communication. However, we will compare hydrocarbon profiles between laboratory-reared and populations of wild-caught flies at several localities. Cuticular lipid amounts will also be compared amongst flies at several localities in Africa..

Development of microsatellite markers for molecular genetic analysis of *Tribolium castaneum* populations in a flour mill.

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The red flour beetle, *Tribolium castaneum*, is a major pest of flour mills. Molecular genetics is a potentially useful tool to address questions about *T. castaneum* population structure, sources of infestation and mechanisms of population rebound after treatment. Beetles for DNA fingerprint analysis were collected in live traps, over the course of several years and from multiple locations within a commercial flour mill. As potential molecular markers, microsatellite regions with polymorphic di- and tri-nucleotide repeat motifs were identified from the literature, BAC-end sequence libraries, and initial data from the genome sequencing project. Polymorphism in these microsatellite markers is being analyzed among individual beetles collected over a range of spatial and temporal scales. To date, at least six polymorphic marker regions have been identified that are detectable on PAGE gels. A hyper-variable region of nuclear DNA was also identified that showed sequence variation among beetles collected within the same mill. These markers will be used to evaluate the amount of gene flow among different subpopulations within a mill and among different mills and grain storage and processing facilities.

Microsatellite markers for the population genetic study of the Indian meal moth, *Plodia interpunctella.*

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The Indian meal moth, *Plodia interpunctella* Hübner, is a serious and widespread pest of stored food commodities throughout the world. The moth is resistant to most of the insecticides used to control it. Proper understanding of the breeding structure, migration, gene flow, and movement of resistant alleles has great implications for management of this pest. Therefore, we have initiated an investigation of the population genetics of Indian meal moth worldwide. As a first step, we developed twelve polymorphic microsatellite markers from genomic DNA enriched for microsatellite loci. Analysis of populations using these loci can give an insight into the population structure, dispersal, genetic relationship and extent of gene flow among the sub populations studied. These markers are an invaluable tool for population genetic studies of Indian meal moth and can be used in DNA fingerprinting for population level identification of this pest.

The mechanism for transmission acquisition of gut symbiotic bacteria in the broad-headed bug *Riptortus clavatus*

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Almost all stinkbugs that feed exclusively on plant juice possess symbiotic bacteria in their midgut. Mechanisms for vertical transmission of the symbiont are impressively diverse among stinkbugs, including egg smearing, ovarial infection, proctophagy, etc. Here we report the discovery of a novel mechanism for transmission and acquisition of the symbiont in the Japanese broad-headed bug *Riptortus clavatus*. *R. clavatus* harbors *Burkholderia* sp. (Proteobacteria; beta-subdivision) as midgut symbiont. However, scanning electron microscopy and diagnostic PCR failed to detect the symbiotic *Burkholderia* from the eggs. Even when juveniles were reared with infected adults in small clean chambers, they did not acquire the symbiont, excluding the involvement of proctophagy. Notably, the symbiotic *Burkholderia* was frequently detected from rhizosphere of the soybean, the main host plant of *R. clavatus*. When hatchlings of *R. clavatus* were reared on potted soybean plants, surprisingly, almost all insects acquired the symbiont. These results strongly suggest that the stinkbug acquires the symbiont not through vertical transmission but from the environment every generation.

Structural analysis of BAC clones containing *Bmper*, *Bmclock*, *T15.180a* and *Rcf96* on the Z chromosome in *Bombyx mori*

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The sex chromosome constitution of the silkworm, *Bombyx mori*, is ZW in the female and ZZ in the male. Until now, the nucleotide sequence of the Z chromosome we determined came up to 1.1 Mb approximately. Construction of a 320-kb BAC contig around the *Bmkettin* gene on the Z chromosome in *Bombyx mori* and determination its nucleotide sequence by the shotgun method were already published. Furthermore, we constructed and determined sequence of a 270-kb BAC contig around the *Rcf96* gene. The nucleotide sequence of three BAC clones, *Bmper*, *Bmclock* and *T15.180a* are about 170-kb each other, were determined in addition, and found 8 novel protein-coding sequences. Analysis of a BAC clone containing *Bmclock* is in progress. All the transposable elements detected in the region were truncated, and no LTR retrotransposons were found, in stark contrast to the situation on the W chromosome. Real-time RT-PCR experiments demonstrated that transcripts of some Z-linked genes were accumulated in larger amounts in males than in female moths. On the other side, transcripts of some genes show larger amounts in females than in male moths, conversely. Consequently, dosage compensation of Z-linked genes was not showed at least in the analyzed region.

Bidirectional cytoplasmic incompatibility with asymmetric partial cross rescue caused by two *Wolbachia* strains in the adzuki bean beetle, *Callosobruchus chinensis*

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Rickettsia-like endosymbiotic bacteria of the genus *Wolbachia* are found in various arthropods. *Wolbachia* endosymbionts are known to induce a phenotype called cytoplasmic incompatibility (CI), one of the selfish strategies of *Wolbachia* to increase their infection frequency in host populations, where crosses between infected males and uninfected females result in lower hatch rates than the reciprocal crosses. We found that the adzuki bean beetle, *Callosobruchus chinensis*, is co-infected with two *Wolbachia* strains, wBruCon (C) and wBruOri (O). To examine whether C and O induce CI or not, mating experiments were performed among four host strains with different infection types: double infection (CO), single infections (C or O) and uninfection (U). C infection and O infection caused strong CI with hatch rates at 0% and 3.3% in incompatible crosses, respectively, while compatible crosses showed 82-92% hatch rates. Notably, while crosses between O females and C males produced no hatched eggs, crosses between C females and O males resulted in 45% hatch. These results could be explained by assuming that C can partially rescue the modification of O. CO females were compatible with males of all infection types, which agreed with the prevalence of CO infection in natural populations.

PCR detection for strand breaks of irradiated plasmid DNA is applied for structural analysis of rDNA cluster including retrotransposons

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"We verified that single strand breaks (SSBs) occurred by gamma ray irradiation were detected by polymerase chain reaction (PCR) method. The frequency of SSBs occurred on both strands of a plasmid DNA region flanked by a pair of primers used for PCR amplifications was calculated by Monte Carlo simulation. The practical data using the PCR reaction with linear shaped pBR322 plasmids irradiated by ^{137}Cs with a Tris-HCl buffer as a radical scavenger, could be almost coincident with patterns on the value, p=0.2, calculated as the case for open circular (OC) plasmids. At the case using closed circular (CC) and OC mixed solution, over 100% values for the non-irradiated mixed solution were observed on low dose condition around 30Gy. This phenomenon was simulated by simple differential equation with possible explanation that CC shape could be very difficult to be amplified by PCR. It is demonstrated that this PCR method using scavengers could also detect a highly complicated DNA structure in water solution. This method should be applied to determination of SSBs frequency for rDNA cluster region including retrotransposons of *Bombyx mori* genomic DNA. We will show the data from the PCR amplification for irradiated genomic DNAs.
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A preliminary molecular phylogenetic investigation of the genera closely related to *Chironomus* Meigen (Diptera: Chironomidae)

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There is considerable uncertainty regarding the true systematic relationships of the genera *Chironomus* Meigen, *Kiefferulus* Goetgh., *Nilodorum* Kieffer, *Einfeldia* Kieffer, *Goeldichironomus* Fittkau. of the Tribe Chironomini of the Chironomidae. Several alternative placements have been suggested by different authors. We have used sequence from the mitochondrial (mt) Cox1 and CytB genes and the nuclear 18S-rDNA (18S) gene, to determine the molecular relationships shown by some members of these genera. The different mutation rates between the mt and the nuclear genes mean that they resolve different levels of the phylogenetic trees: Mt sequences resolve relationships at the tips of the branches, while 18S sequences resolve the deeper branches of the trees. By using both data sets we can obtain a better evaluation of the overall relationships within and between genera. These preliminary results support the inclusion of *Nilodorum* in the genus *Kiefferulus,* as well as the placement of *Kiefferulus* as sister genus to *Chironomus.* They further suggest that *Einfeldia dissidens* Walker is close to *Chironomus*, but distinct from *C. javanus* Kieffer, which was at one time considered to bean *Einfeldia* species, but which clusters here within *Chironomus,* as is more generally assumed.

The delay of mating time through long term mass-rearing in the melon fly, *Bactrocera cucurbitae* (Coquillett)

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The eradication of the melon fly, *Bactrocera cucurbitae* (Coquillett) from Okinawa Japan using the Sterile Insect Technique (SIT) was achieved in 1993. In the eradication program, three different mass-reared strains, named K strain, M strain and OY strain, were used for control in Kume Island, Miyako Islands and Okinawa/Yaeyama Islands, respectively. To prevent re-colonization by alien flies, OY strain is still culturing continuously and being released in the Ryukyu Islands. This strain has been reared for ca. 150 generations under artificial conditions. So we concern for loss of control efficiency in SIT because of the flies being domesticated. Herein, we compared the temporal changes of the mating behavior between Taiwan wild and OY strains of melon fly in order to examine whether the differences of the mating traits exist between them or not. Moreover, in order to estimate the changes of mating traits through the long term rearing, we also compared them between OY strain and K strain which has been reared continuously in small-scale for ca. 215 generations. We present the results of these tests and consider the serious changes which may occur through the long term mass-rearing.

Comparison of life history characters of arrhenotokous and *Wolbachia*-associated thelytokous *Trichogramma kaykai* Pinto & Stouthamer (Hymenoptera: Trichogrammatidae)

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We analyzed the life history characters related to rate of potential population growth for two genetically identical lines of *Trichogramma kaykai* Pinto and Stouthamer. These lines differed in that one was infected with parthenogenesis-inducing *Wolbachia* (W^+), while the other was free of *Wolbachia* (W^-) through antibiotic treatment. For both lines the number of females emerging per day was the highest from eggs laid during the first two days of oviposition. Although adult longevity in W^- was shorter than that in W^+, number of eggs reaching the prepupal stage, adult emergence, and female emergence were significantly higher for W^- than for W^+. There were no significant differences in the number of mature ovarian eggs at emergence between W^- and W^+. The values of mean length of generation (T) in W^+ was similar to that in W^-, but the values of net reproduction rate (R_0) and intrinsic rate of natural increase (r_m) in W^+ were lower than those in W^-. In most field populations, the W^+ individuals form a small percentage (<5%) of the total population. The relative low growth rate caused by the *Wolbachia* infection may contribute to this low prevalence of the infection. Because differences in the relative population growth rates would soon result in the extinction of the infected population in the field, other factors must play a role allowing the infection to maintain itself in the field.

Expressed sequence tag (EST) approach to understand the host role in aphid bacteriocyte symbiosis

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Almost all aphid species (Homoptera, Aphididae) have bacteriocytes that are specifically differentiated to contain endocellular symbiotic bacteria of the genus *Buchnera*. Since the initial infection (150-250 million years ago), *Buchnera* have been vertically transmitted to eggs and embryos through host generations, and the mutualism between the host and *Buchnera* is so obligate that neither can reproduce independently. Complete genome sequencing of *Buchnera* from the pea aphid, *Acyrthosiphon pisum*, revealed that the genome is so small (640,681-bp chromosome and two small plasmids) that it lacks many genes that appear to be essential to bacterial function, whereas it retains genes for the biosyntheses of nutrients that are required by the host aphid. Our main focus is on the host's role in maintaining this symbiotic system. In an effort to understand this, we constructed a cDNA library of the host bacteriocyte using Cap-trapper method, and are performing expressed sequence tag (EST) analyses. So far, we have sequenced approximately 3,000 clones, among which BLAST similarity searches and subsequent real-time quantitative RT-PCR analyses detected a number of candidate gene products that may be essential to the symbiotic system.

Wolbachia infection and mt-DNA of the pierid butterfly, *Eurema hecabe*

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Eurema hecabe (L.) thrives in the Oriental tropics as well as the subtropical to temperate zones. In Japan, the population in the subtropical southwestern islands differs from that in the temperate mainland in terms of the seasonal form of adult wing color pattern expression and larval food habits. We analyzed allozyme variation in populations from the mainland and the southwestern islands. Zymograms and allele frequencies differed between the two populations. Allozyme data of individuals with yellow fringe color of the forewing (yellow type) were also different from those of brown butterflies distributed sympatrically in Okinawa Island. In the present study, we used the DNA sequence data of the mitochondrial ND5 gene to clarify the phylogenetic relationship between the two types of *E. hecabe* Recently, it was reported that *Wolbachia* – a kind of rickettsia– could infect the cytoplasm of *E. hecabe.* We examined all individuals using molecular phylogenetic methods to determine infection with *Wolbachia.* It is surprising that of the two separate groups in the phylogenetic tree, one group consisted of *Wolbachia*-infected individuals and the other consisted of non-infected ones. It is therefore suggested that the mtDNA of *E. hecabe* is not a useful tool for molecular phylogenetic analyses.

Molecular cloning and expression analysis of the Bmfru gene, the *Bombyx* homologue of the *Drosophila* sex determination gene *fruitless*

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The *Drosophila fruitless* (*fru*) gene has a key role in the sex determining cascade. The *fru* regulates male sexual behavior and motorneuron-dependent formation of the male specific abdominal muscle of Lowrence (MOL) through its functioning in the nervous system. We have isolated a homologue of *fru* in the *Bombyx mori*, which is referred as *Bmfru*. Four types of cDNA clones were isolated from cDNA libraries. All of these transcripts encodes proteins related to the BTB-ZF protein family. The BTB domain of *Bmfru* showed 91% of identity with that of *D. melanogaster*. Three of four BmFRU isoforms have variations in their carboxy terminus in which zinc finger motifs are contained. The *Bmfru* mRNAs are mainly expressed in gonads and heads of 5th inster larvae and pupae. From the results of in situ hybridization and immunohistochmical analysis, it is revealed that *Bmfru* mRNAs and proteins are expressed in gonadal epithelium, optic lobe and suboesophageal ganglion.

Mating interactions between the okra and cassava biotypes of *Bemisia tabaci* (Gennadius) (Homoptera: Aleyrodidae) on eggplant

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Bemisia tabaci is an important pest of field and horticultural crops worldwide. It displays wide host-spectrum, diverse types of crop damage and intraspecific variation, reported in the existence of over 20 biotypes. Genetic and biological variation within *B. tabaci* has great implications on its pest status, control and the integrity of the species as a single taxon. Reproductive isolation between biotypes has been demonstrated and perhaps illustrates the genetic relationships between biotypes, apart from influencing biotype interactions in nature. In this study, the biological consequences of mating between okra and cassava biotypes were assayed in clip cages on eggplant, by reciprocal crossing. Interbreeding had no significant influence on the fecundity of the mated females and survival of progeny (P.0.05). Female crosses were not morphologically different from purebred females ($p>0.05$), and oviposited normally. However, the sex ratio of hybrid progeny was significantly male dominated (<30 % females) compared to the pure bred progeny of each biotype. (>60% females) ($p < 0.01$). Therefore, there seems to be limited possibility of gene flow between the two biotypes on shared hosts. Still, some reproductive incompatibility exists, and perhaps buttresses ecological isolation in nature, hence maintains the genetic and biological identity of the two biotypes.

Rapid cold hardening increases aphid survival at low temperatures

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Most studies on insect cold hardiness have focused on seasonal adaptations occurring within the overwintering stage of an insect's life cycle, triggered by decreasing temperature and photoperiod from summer through autumn to winter. Due to their short generation times, an aphid that enters winter is unlikely to be the same individual that survives until spring. For this reason, although aphids are able to cold harden when reared at lower temperatures (10° and 5°C), the ability to rapidly cold harden may be relatively more important than in other longer-lived overwintering insects, where seasonally-induced cold hardening is the dominant response. This study assesses the effects of sub-zero temperatures on the mortality of nymphs and adult *S. avenae*, provides evidence of rapid cold hardening in *S. avenae* and investigates the ecological costs of rapid cold hardening in terms of effects on development, longevity and fecundity. The hypothesis that rapid cold hardening can be induced during the cooling phase of natural diurnal temperature cycles is supported. Rapid cold hardening increases survival and extends the lower lethal temperature, allowing aphids to acquire maximum cold hardiness when it is most needed.

Wolbachia **replacement in global populations of** *Drosophila melanogaster*

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Wolbachia are endosymbiotic bacteria associated with a large variety of arthropod species. They are mainly inherited vertically and have successful strategies to colonise host populations by manipulating host reproduction. Recently the genome of the *Wolbachia* strain infecting *Drosophila melanogaster* has been sequenced. The small genome is unique for its large number of repetitive elements and also contains potential sites for variable number tandem repeats (VNTR). Analysis of *Wolbachia* in globally representative and historic populations of *D. melanogaster*, kept at several stock centres, revealed polymorphisms in repetitive element sites and VNTR sites, indicating the presence of two related but distinct *Wolbachia* strains in this host species. The distribution of the two strains is strongly correlated with the collection dates of the fly lines. The results strongly suggest that there has been a historic replacement of one *Wolbachia* strain by another, probably caused by different strengths of host reproductive manipulations. It is intriguing that the sweep of the present *Wolbachia* strain occurred in the period of the colonisation of the *D. melanogaster* genome by P elements.

Comparative analysis of *Anastrepha sororcula* (Diptera: Tephritidae) populations based on rDNA and karyotypes

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The true-fruit-fly *Anastrepha sororcula* is an important Brazilian fructicultural pest, since it attacks several commercial valuable fruits. Samples of *A. sororcula* from the Brazilian States, São Paulo (SP), Goiás (GO), Bahia (BA) and Rio Grande do Norte (RN), were submitted to a comparative study based on their karyotypes and ribosomal DNA. Mitotic metaphasic chromosomes were analysed after DAPI coloration. Number (2n=12) and sizes of chromosomes in all samples were similar, except SP sample which showed distinctive DAPI staining pattern. X chromosomes of SP sample showed a submedian interstitial and a pericentromeric DAPI+ band, whereas samples from GO, BA, RN presented only the submedian interstitial band. All populations exhibited autosomes with tiny pericentromeric DAPI+ bands and a dry chromosomes entirely heterochromatic. Differences among samples from SP, GO, BA, RN regarding rDNA were detected in Southern blots. DNA of males and females was cleaved with endonuclease *Hin*dIII and hybridised to rDNA 45S probe. The four samples were distinguished by hybridisation pattern although each sample was monomorphic. Hitherto, partial sequencing of 300 bp of 3´-end of ITS1 showed no differences among the samples. Results indicated genetic differences among geographically distant populations of *A. sororcula*, suggesting that this generalist species present low dispersion behaviour.

Value of the morphometric study in the discrimination of two species of the *flavida* complex (Hemiptera: Reduviidae).

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The flavida complex, restricted to the Antillas, is composed of three related species: *Triatoma flavida, T. bruneri* and *T.obscura*. *T. flavida* and *T. bruneri* are species endemics from Cuba and *T. obscura* from Jamaica. The taxonomic status of *T. flavida* and *T. bruneri* has been very discussed by their similarity morfhological. We presented here, for the first time, a study morphometric comparative between these species using an discriminant analysis included and excluded size using the PADWIN program (Dujardin, 2002). We used characters of the head, thorax and wings for the analysis whrere the data were previous transformed to logarithms. The multivariate analysis showed broadly quantitative differences between both species, confirming the value of the morphometry in order to distinguish species closely related.

Phylogenetic characterization of primary symbionts associated with diverse whitefly species.

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Many insects, including whiteflies, have intracellular microorganisms that are considered symbiotic in nature. Primary endosymbiotic bacteria, housed in specialized cells of insect origin called mycetocytes or bacteriocytes, have primarily been associated with nutritional functions by providing essential nutrients to the insect. Although symbionts of whiteflies have been studied, those data focus primarily on only *Bemisia tabaci* biotypes and haplotypes. Little is known about the composition and diversity of the endosymbionts in other Aleyrodidae. We have examined over 50 different species of whiteflies by PCR amplification and subsequent sequencing of the products produced from primary symbiont 16S ribosomal DNA (rDNA) genes directly from insect tissue. Maximum parsimony analysis and congruence between the phylogeny of the symbionts and their insect hosts were examined. In addition, bacterial endosymbionts of several species were morphologically characterized to determine the relative number of organisms, the presence or absence of a cell wall, and the nature of the cell membranes. Variation in these characteristics, and diversity or abundance of organisms in the bacteriomes and bacteriocytes will be discussed.

Population variation in striped rice borer, *Chilo suppressalis* Walker (Lep.: Pyralidae) using geometric morphometrics

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The morphological variation of a total of 311 forewings and 319 hind wings of the Striped Rice Borer collected from 6 different geographic localities in Iran were analysed using geometric morphometric techniques. 24 & 14 partial wrap scores and 2 & 2 uniform components calculated for each specimen in forewings and hind wings respectively and saved as a Weight Matrix using tpsRelw. In forewings and hindwings respectively 9 & 6 components extracted with eigenvalues greater than 1. Totally they accounted for 71.43% of variation were in forewings & 71.37% in hindwings. Amongst the 281 observations used to fit the model, 158 or 56.23% were correctly classified. Graphical options represented that the western geographical populations differ from eastern populations, also two near localities named Rasht and Anzali showed degrees of differentiation between them. Then performed DFA for hindwings and the results showed that amongst 300 observations used to fit the model, 129 or 43.8% were correctly classified. Graphical options represent the same results like forewings, also Anzali population showed very clearly separation among other populations. The results of Cluster analyses on the mean of populations and the dendrogram confirmed results.

Genetic factors affecting egg size in a migrant skipper, *Parnara guttata guttata* (Lepidoptera: Hesperiidae): genetic relation with life history traits and variation in pattern of reaction norms

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Genetic factors controlling egg size were examined in the migrant skipper, *Parnara guttata guttata* to detect the genetic basis of egg size variation which is needed for a discussion of the evolution of egg size. First, phenotypic and genetic correlations and heritabilities were estimated for egg size, fecundity and body size (forewing length) under the different day-length conditions (16L8D and 14L10D at 25 ^{0}C). Second, genotype-environment interactions were estimated by comparing reaction norms to day-length of these traits among families. Negative phenotypic and genetic correlations between egg size and fecundity were estimated in both environments. High negative correlations were found between body size and egg size under 14L10D at phenotypic and genetic levels. Narrow sense heritabilities estimated by parent-offspring regression were significantly differed from zero for egg size and fecundity. These results show that egg size varies genetically and it can be affected by the genetic relation with other life history traits. Two-way ANOVAs showed significant genotype-environment interactions in egg size. The variation observed in phenotypic plasticity of egg size is discussed in relation to optimal allocation strategy for energies to produce eggs and seasonal migration strategy in *P. g. guttata*.

Occurrence of nematode inside the malpighian tubules of *Hypocryphalus mangiferae* (Coleoptera: Scolytidae).

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Hypocryphalus mangiferae is considered the vector of the fungus *Ceratocystis fimbriata* from mango tree. As part of an ongoing study to locate the presence of a mycangium in the body of this beetle, we found the occurrence in all 50 dissected specimens of a dilated region in one Malpighian tubule containing great amounts of nematodes. In freshly dissected beetles was possible to verify the movement of nematode towards the hindgut of beetle. This structure was found in both sexes and apparently the beetles were not affected because they were feeding and reproductive as determined by histological analysis of digestive and reproductive tracts. The structure containing nematodes were always in the same Malpighian tubule, which was characterized by the flattening of its epithelium in comparison with Malpighian tubules without nematodes. We suggest a phoretic association between the nematode and the beetle. The identification of the nematode species remains to be determined.

Analysis of population structure of *Bactrothrips brevitubus* (Thysanoptera : Phlaeothripidae) using DNAmicrosatellite marker

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Bactrothrips brevitubus is haplodiploid, facultatively ovoviviparous, Japanese thrips. Females can control the sex of their offspring. Individual females can produce both sexes in a mass of eggs oviparously and only males by single eggs ovoviviparously. Sex ratios are split according to the dominant reproductive mode within a patch (Kranz et al. 2002). Local mate competition (LMC), local resource competition (LRC) for food and egg laying sties, and parasites are possible factors influencing reproductive mode. In this study, we tested for LMC by estimating relatedness in patches using microsatellite DNA. Microsatellites also permit detailed analyses of the local population structure. For primer development, we used the dual-supression-PCR technique of Lian et al. (2002). This technique is simple and effective for organisms with a low frequency of microsatellites. Our microsatelite data for gene frequency and relatedness indicate that individuals disperse between patches but not between populations, providing support for our hypothesis that LMC influences sex allocation and reproductive mode in these thrips. (160 words)

Male-male mounting behavior in *Ptomascopus morio* (Coleoptera: Silphidae)

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Ptomascopus, a sister genus of *Nicrophorus*, uses small vertebrate carrion as food for their young but shows no parental behavior like *Nicrophorus*. When we investigated sexual behavior of *Ptomascopus morio,* we found curious behavior which male mount another male like copulation. In some trials, the mounting male extended his aedeagus in the copulatory orientation (facing forward). We call this behavior as mounting, and the level of male-male mounting was compared in the presence of two different stimuli (a large female versus a resource for breeding). In almost all cases the larger male mounted the smaller male. Number of copulation was not different whether carrion was present or absent. However, presence of carrion elicited more aggressive acts and mounting than absence of carrion. Thus we hypothesize that male-male mounting behavior is not misidentified males as females, but reflects aggression in some cases.

The isolation mechanism of *Drosophila albomicans* & *D. nasuta*

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Drosophila albomicans and *D. nasuta* show a little but not significant postmating reproductive isolation. However, no hybrids have ever been found in nature anywhere including the area near the border according to their distinct karyotypes (2n=6 in *D. albomicans* and 2n=8 in *D. nasuta*). Studying the cause of preventing gene flow between them is important to realize population differentiation as well as speciation. Previously data showed the survival and fecundity of hybrids have no significant decrease, so we hypothesized the prezygotic isolation is the main events to prevent the gene flow in nature of this two species. Prezygotic mechanism was further divided into two periods: premating and postmating-prezygotic. The former was analyzed by the cage experiments of multiple-choice mating, further detect whether the *D. albomicans* and *D. nasuta* of different geographic regions have acquired different prezygotic reproductive isolation mechanisms.

Host plant specialisation governed by facultative endosymbiont

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Host plant specialization of herbivorous insects has fascinated many entomologists as well as evolutionary biologists. Traditionally, the ecological trait has been attributed to genotype of the insects themselves, but here we found that plant adaptation of the pea aphid, *Acyrthosiphon pisum*, is strongly affected by a facultative endosymbiotic bacterium. Almost all aphids harbor an essential endosymbiotic bacterium, *Buchnera* sp.. In addition, many aphids possess facultative endosymbiotic bacteria, which are collectively called secondary symbionts. At three locations in central Japan, occurrence of a secondary symbiont, called pea aphid U-type symbiont (PAUS), was consistently higher on white clover than on vetch. By an antibiotic treatment, we created genetically identical aphid strains with and without PAUS infection. Whereas these strains exhibited similar levels of fecundity on vetch, the PAUS-eliminated strain lost almost 50% fecundity compared with the infected strain on white clover. When PAUS was reintroduced into the eliminated strain, the reduced fecundity on white clover recovered almost completely. These results indicate that PAUS infection improves the fitness of the host aphid specifically on white clover. This finding would shed new light on some aspects in evolutionary ecology of aphids and other herbivores, including host race, ecotype, and sympatric speciation.

Identification of primary tissues showing the lowest hardiness to freezing in larvae of the rice stem borer, *Chilo suppressalis* Walker (Lepidoptera: Pyralidae)

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Even though overwintering larvae of the rice stem borer, *Chilo suppressalis* are freeze-tolerant; they can't survive below -30°C. Furthermore, non-diapausing larvae can't survive freezing. However, the cause of freezing death is unclear. To identify the cause of freezing-death of larvae, we tried to identify the primary tissues which were injured by low temperatures. Trypan blue dye was used to determine the injured tissues. In overwintering larvae, the midgut and silk glands of dead larvae were stained blue, and remarkable color density differences between dead and surviving larvae were observed in the midgut. In non-diapausing larvae incubated at -10°C for several hours, the fat body of dead larvae was strongly stained. Furthermore, increases in mortality with treatment time for larvae corresponded with increases in the area of the fat body stained. Sterile non-diapausing larvae with a lower supercooling point of below -20°C did not freeze at -10°C and survived the treatment. However, all the larvae died when subjected to inoculative freezing at -10°C, and the fat body was stained blue. These results suggest that the midgut in overwintering larvae and the fat body in non-diapausing larvae have the lowest hardiness to freezing.

Identification and sequence evolution of the gene encoding antimicrobial peptide defensin in ants

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Organisms that live in social groups have greater susceptibility to infectious diseases compared to solitary organisms. In social insects the interaction with pathogens has led to a number of adaptations to reduce their deleterious effects. Such adaptations may be revealed by studying the existing diversity and long-term evolution of genes involved in insect immune system. One component of insect innate immune system, the humoral response, is triggered by the recognition of invading microorganisms resulting in the production of antimicrobial peptides. These peptides, and the microbes they target, may coevolve. In the nucleotide level this might be seen as directional selection or selection maintaining polymorphism. I have cloned and characterized the gene coding the antimicrobial peptide defensin from the ant *Formica aquilonia*. Sequence comparisons of several related ant species reveal that the gene is conservative in the group of species studied. The next step is to expand the comparison to phylogenetically more remote species and to other genes of the ant immune system.

Links between cold hardiness and desiccation resistance in an overwintering insect

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To investigate possible links between cold-hardiness and desiccation resistance in insects, we examined changes in seasonal parameters associated with these phenomena in overwintering larvae of the goldenrod gall fly *Eurosta solidaginis.* During the autumn, field-collected larvae gradually increased their capacity to survive freezing concomitantly with increases in cryoprotectants as measured by hemolymph osmolality (488 to 870 mOsm•Kg^{-1}). In contrast to gradual increases in larval cold hardiness, resistance to desiccation increased in two distinct phases. An initial 6-fold decrease in rates of water loss occurred in a two-week period in early October, reaching 0.57 μg•mm^{-2}•h^{-1}; this phase was primarily due to a reduction in respiratory water loss as the larvae entered diapause. The second phase was a more subtle 4-fold reduction that occurred over eight weeks beginning in mid-October. Interestingly, the second phase of reduced rates of water loss correlated strongly with increased hemolymph osmolality (568 to 870 mOsm•Kg^{-1}). In addition, we acclimated larvae at various temperatures in order to manipulate cryoprotectant concentrations. Our preliminary results suggest a trend exists between higher hemolymph osmolalities and lower rates of water loss. Taken together these data suggest that cryoprotectants may promote desiccation resistance as well as cold-hardiness in overwintering insects. Supported by NSF#IBN-0090204.

Species-diagnostic traits unravel under the heat: Hybrid zone dynamics and cryptic mountain species revealed by changing thermal landscapes

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Recently (and probably directly related to extensive regional global warming in the region since 1998) considerable northward and rapid interspecific introgression (150-350 km in 6 years) into historical *P. canadensis* populations has been observed for several autosomal species-diagnostic traits such as detoxification of tulip tree leaves, morphological wing traits, and *Hk*-100 allozymes . In addition, some X-linked traits such as *Pgd*-100 alleles and tulip tree oviposition preference behavior have moved extensively northward. However, there has been little introgression of certain other sex-linked *P. glaucus* traits (such as facultative diapause, dark morph females, and LDH-100 allozymes). Recombination and strong selection on some undetermined trait (and close linkage of *Ldh*-100 with that trait on the X-chromosome) seem to limit gene flow North of this interspecific hybrid zone whereever fewer than 2700 degree-days accrue. We have created recombined/introgressed hybrids that largely match the morphological and delayed post-diapause emergences of the new mountain swallowtail species, *P. appalachiensis* (Pavulaan & Wright 2002). In the lab, diapausing backcrossed hybrids are delayed in post-diapause spring emergences by 2-3 weeks after the parental species. The new species expresses an introgressed mix of both *glaucus* and *canadensis*. diagnostic traits and may represent the high altitude version of our high latitude northern "false second generation" populations. GIS mapping of thermal accumulations allows prediction of the geographic location of the purportedly new species *P. apppalachiensis* (Pavulaan and Wright 2002) in West Virginia as well as for Georgia in the southern Appalachian Mts

Evolutionary trade-offs between reproduction and dispersal in populations at expanding range boundaries

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During recent climate warming, some species have expanded their ranges northwards to keep track of climate changes. Evolutionary changes in dispersal have been demonstrated in these expanding populations and here we show that increased dispersal is associated with reduced investment in reproduction in populations of the speckled wood butterfly, *Pararge aegeri*a. Evolutionary changes in flight versus reproduction will affect the pattern and rate of expansion at range boundaries in the future, and understanding these responses will be crucial for predicting the distribution of species in the future as climates continue to warm.

Cold tolerance and feeding behaviour in the Tasmanian scorpionfly *Apteropanorpa* Carpenter (Mecoptera: Apteropanorpidae)

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The Apteropanorpidae is a small family of scorpion flies endemic to Tasmania, comprising two described species. The biology of the family is almost completely unknown. Adults of both species are found on shrubs in the alpine zone and can be active in winter, although the peak in adult abundance is autumn. Experiments cooling individuals from 3°C to −10°C at 1°C/3 mins and measuring body temperature using infrared video thermography show that live *Apteropanorpa* freeze at an average temperature of −4.9°C (n=20). However, stress response behaviour is triggered at an average temperature of −1.1°C (n=20), and no individuals survived freezing to −10°C. Combined behavioural and physiological data indicate that *Apteropanorpa* does not possess a physiological mechanism to survive freezing, and instead employs freeze avoidance behaviour as the insect seeks microhabitats with favourable temperatures. *Apteropanorpa* is apparently well-adapted to its natural environment; the maritime climate of the Tasmanian alpine zone is relatively warm and stable compared to that of mainland Australia. Adults are most likely saprophagous in nature, predominantly feeding on dead and decaying invertebrates. Thirty-three adults were included in a feeding choice experiment in which twenty-two adults showed a statistically significant preference for feeding on dead arthropods. A saprophagous feeding strategy in the Apteropanorpidae is supported by field observations, histological tests and gut contents analyses by light and electron microscopy.

Jumping Genes and AFLP Maps: Transforming Lepidopteran Color Pattern Genetics

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The color patterns on the wings of Lepidopterans are among the most striking patterns in nature and have inspired diverse biological hypotheses such as the ecological role of aposomatic coloration, the evolution of mimicry, the role of human activities in industrial melanism, and the developmental basis of phenotypic plasticity. Yet, the developmental mechanisms underlying color pattern development are not well understood for three reasons. First, few mutations that alter color patterns have been characterized at the molecular level, so there is little mechanistic understanding of how mutant phenotypes are produced. Second, while gene expression patterns resembling adult color patterns are suggestive, there is little data available showing that gene products have a functional role in color pattern formation. Finally, because with few exceptions (notably *Bombyx*), genetic maps for most species of Lepidoptera are rudimentary or nonexistent, making it very difficult to determine whether mutations with similar phenotypes are due to lesions in the same gene or different genes. This presentation will show how a combination of germ line transformation using transposon vectors and genetic mapping employing variation between closely related species is rapidly resolving these difficulties and providing much new information about the development and evolution of these color patterns.

CARDINIUM HERTIGII, A NEWLY DISCOVERED SYMBIONT IN THE *BACTEROIDETES* CAUSES MULTIPLE REPRODUCTIVE MANIPULATIONS OF ITS HOSTS

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Vertically transmitted symbionts of arthropods have been implicated in several reproductive manipulations of their hosts, including cytoplasmic incompatibility (CI), parthenogenesis induction in haplodiploid species (PI), feminization, and male killing. *Wolbachia*, an a-proteobacterium is the only symbiont known to cause all of these effects, and has been thought to be unique in causing parthenogenesis and CI. We found a symbiont lineage in the *Bacteroidetes* group, recently described as *Cardinium hertigii*, causes PI and CI in parasitoids in the genus *Encarsia. Cardinium* is strongly associated with parthenogenesis in *Encarsia*, and in *E. hispida,* observations indicate it is the causative agent. In a sexual population of *E. pergandiella*, *Cardinium* induces CI. The 16S rDNA sequence of *Cardinium* in *E. pergandiella* is 99% identical to a parthenogenesis-inducing strain in another population of *E. pergandiella,* indicating that like *Wolbachia,* closely related strains can cause different phenotypes. *Cardinium* in *Encarsia* is also 96% identical to a feminizing symbiont in haplodiploid *Brevipalpus* mites. Thus, *Cardinium* has been found to induce three of the four principal manipulations of host reproduction known from *Wolbachia*. The discovery of *Cardinium* permits comparative analyses not possible when *Wolbachia* was thought to be unique.

The reproductive effects of infection by the intracellular bacterium *Cardinium* in mites

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The endosymbiont *Wolbachia* has received widespread notoriety over the past decade because of its high infection frequency among arthropods, and the unique heterogeneity of reproductive effects it has been implicated as causing to enhance its own spread. However, the newly described intracellular bacterium *Cardinium* has recently been implicated as causing a variety of reproductive manipulations in its host's that rivals *Wolbachia* for overall diversity. In addition, a recent screening study showed that a significant number of arthropods are infected with *Cardinium*, with the greatest number of infected species found in the Acari (mites). Here, I will talk about the reproductive effects induced by *Cardinium* in several different groups of mites, including feminization in *Brevipalpus* mites, fecundity enhancement in the predatory mite *Metaseiulus occidentalis* and parthenogenesis induction in several oribatid mite species. Finally, I will comment on the presence of double infections of *Wolbachia* and *Cardinium* in several host species and the implications of these infections for determining effects caused by these intracellular microorganisms.

Biological interplay between aphid, *Buchnera*, and facultative endosymbionts: importance of unrecognized players

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Almost all aphids harbor the essential endosymbiotic bacterium *Buchnera*. In addition, many aphids possess additional endosymbiotic associates collective called "secondary symbionts". On the basis of physiological roles such as supply of essential amino acids, *Buchnera* is indispensable for life and reproduction of aphids. Although neglected for a long time, secondary symbionts have recently attracted much attention from researchers, and intensively investigated by using new techniques such as molecular phylogenetics, *in situ* hybridization, quantitative PCR, and artificial transfer and selective elimination, which shed new light on a variety of important biological roles carried by the unrecognized players. Here we report our new findings on the biology of secondary symbionts in aphids. 1) In Japanese pea aphids, a facultative symbiont called PAUS is involved in host plant specialization. 2) A facultative symbiont called PASS can support survival and reproduction of *Buchnera*-free pea aphids that will otherwise be completely sterile. 3) In some aphid groups like *Yamatocallis* spp. and *Colophina* spp., secondary symbionts are not facultative associates but are found from all individuals and populations of related species, suggesting that these secondary symbionts may play some important roles for the hosts and consequently be essential.

Theoretical and empirical studies of *Wolbachia* infections in mosquitoes

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Recent progress characterizing *Wolbachia* infections in medically important mosquito populations will be presented. *Wolbachia* is an obligate intracellular infection of invertebrates that is maternally transmitted through the host embryonic cytoplasm. *Wolbachia* can cause cytoplasmic incompatibility, which effectively penalizes females that lack infection by reducing their fecundity. In natural populations, cytoplasmic incompatibility can promote the spread and maintenance of *Wolbachia* infections. Artificial manipulation of *Wolbachia* infections and cytoplasmic incompatibility can be used in applied strategies for the suppression of economically important pest populations and for 'population replacement' of medically important disease vectors. The presentation will include recent characterization of *Wolbachia* infections in *Aedes* and *Culex* mosquitoes, research developing new molecular markers for discriminating between *Wolbachia* infections in the *Culex pipiens* complex, theoretical and empirical examination of *Wolbachia* effects on *Aedes albopictus* populations, and a new model for the evolution of new cytoplasmic incompatibility types.

The genetic basis of variation in butterfly wing patterns: Evolution, and development in *Bicyclus anynana*.

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Heritable phenotypic variation is the raw material of adaptive evolution, and understanding its generation is a crucial issue in contemporary evolutionary biology. Furthermore, it is becoming widely recognized that variation should be of focal interest across all disciplines in biology. Genetic and phenotypic heterogeneity are a universal characteristic of living organisms, with variation across- and within-species existing for essentially all phenotypes. Studies in insects have made important technical and conceptual contributions to our understanding of phenotypic variation and its basis. Wing patterns of *Bicyclus* butterflies, in particular, have proven ideally suited to study the reciprocal interactions between evolutionary and developmental processes that shape variation within and across species for a morphological phenotype of ecological relevance. There is much variation in wing patterns in *B. anynana*, and detailed studies of the developmental pathways leading to pattern formation. To link these sets of data we need to know which genes within the implicated pathways contribute to variation in wing patterns in *B. anynana* and related species. Results of experiments exploring variation in wing patterns will be discussed, and so will our efforts to develop the genomic tools with which to dissect its genetic and developmental basis.

Sexual selection shapes genital morphology in a water strider: blurring the distinction between primary and secondary sexual traits.

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Recent evidence that insect genitalia are subject to sexual selection has blurred the distinction between primary and secondary sexual characteristics. However, the extent to which genital morphology has been shaped by sexual selection remains controversial, as does the primary mode through which sexual selection acts on genital form and function. To discern the role of sexual selection in shaping genital evolution, we present a case study of the water strider, *Aquarius remigis* (Hemiptera, Gerridae), a species characterized by unique genital morphology, a polygynandrous mating system, and active female resistance to mating attempts. Our field assays demonstrate that sexual selection acting through differential mating success consistently favors males with long external genitalia, most probably because long external genitalia provide mechanical advantage to males as they try to position and insert the phallus. Video and microscopic studies of intromission, phallus inflation and insemination suggest that several features of the inflated phallus also enhance male paternity success more cryptically by anchoring the male in copula and by displacing and damaging sperm from previous males. Our results thus support the hypothesis that the unusual genital morphology of this species has evolved in response to sexual selection acting through both overt and covert intrasexual selection.

Genital variation in a dimorphic moth *Selenia tetralunaria*

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It is generally assumed that genitals do not vary within a species. We inspected morphologies of male and female genitalia of bivoltine geometrid moth *Selenia tetralunaria*. The two annual generations differ in morphology so that the second generation individuals are approximately 20% smaller than those of the first generation, and they also differ in wing patterns and shape. The morphs do occasionally co-occur. We found that that as reported in many insect and spider species genitalia show negative allometry in relation to body size. We found that this allometry is stronger in internal genital structures than in external genitalia. We also found that there is more variation in internal than in external genitalia. Thirdly, we found that the shape of genital structures differ between morphs in all three examined areas. *S. tetralunaria* is among the first reported cases of genitally dimorphic insect species. Considerable variation in internal genitalia and especially the presence of genital shape differences between morphs are not consistent with the predictions of lock-and-key hypothesis.

Genetic differentiation in three *Cryptophlebia* (Lepidoptera: Tortricidae) species of economic importance in South Africa

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The false codling moth, *Cryptophlebia leucotreta,* is one of the most damaging and catholic insect pests in South Africa. The pattern of genetic differentiation was studied in this species and compared with those of the litchi moth *C. peltastica* and the macadamia nut borer *C. batrachopa*. Populations of all three species were collected throughout their known range in South Africa from a variety of tropical, subtropical and deciduous hosts. Populations were studied using amplified fragment length polymorphism (AFLP) analysis and nucleotide sequence variation of the mitochondrial cytochrome oxidase 1 (COI) gene. Significant intraspecific variation was detected in all three species although *C. leucotreta* appeared as the most variable species, while *C. batrachopa* was the least variable. Analysis of molecular variance (AMOVA) revealed a high level of variation within populations for all three species. Estimates of the absolute number of migrants between populations (Nm) based on data obtained from mitochondrial and nuclear DNA markers indicate that *C. leucotreta* has a strong geographical structure with low gene flow between geographic populations. However, populations collected from different hosts differed only slightly. Furthermore, evidence was found for gender related dispersal. The results obtained are explained and used to provide insight into managing population levels of *Crytophlebia* spp. in different environments.

Population structure of *Plutella xylostella* (L.) investigated using microsatellite markers

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Diamondback moth (*Plutella xylostella*) is found throughout Australia infesting *Brassica* vegetables, canola, forage brassicas and weeds, but its long-range movement patterns in this country have not been ascertained. The species is considered highly migratory in the Northern Hemisphere and is known to be capable of travelling long distances on high altitude winds. Management of the pest in Australia and around the world would be facilitated by knowledge of its population genetic structure and dispersal patterns. We have developed six polymorphic microsatellite markers from a partial genomic library and an EST library of diamondback moth and are using them to investigate these factors. Populations of diamondback moth from around Australia will be compared with populations from Africa and Asia. Preliminary analysis of the Australian data indicates little genetic differentiation between diamondback moth populations within the country. However, a population from Kenya is showing a higher degree of microsatellite allele polymorphism than the Australian samples.

Drought responsive genes in the sleeping chironomid, *Polypedilum vanderplanki*

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"Organisms have evolved various mechanisms for adaptation to adverse environmental conditions. The sleeping chironomid, *Polypedilum vanderplanki*, is the largest multicellular animal known to tolerant almost complete dehydration without ill effect. The anhydrobiotic (cryptobiotic) larvae could survive at extreme temperatures from -270°C to +106°C (Hinton, 1968). We have demonstrated that rapid accumulation of trehalose coincided with loss of water form the larvae, also that cerebral regulation was not involved in the process of anhydrobiosis like plants or unicellular organisms. To address molecular events during desiccation going into anhydrobiosis in this insect, we cloned several genes concerning trehalose metabolism, water transport and so on by RT-PCR using degenerated primers and screening EST database of *P. vanderplanki*, and subsequently carried out Northern blot analysis to measure amounts of expressions of these genes during desiccation. Consequently, we obtained five drought responsive genes at least. We will discuss how these drought responsive genes orchestrate anhydrobiosis in *P. vanderplanki*.
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Ecological adaptation of circadian activity rhythms: phase shift and loss of rhythmicity with the altitude change in Plusinae (Noctuidae) moths

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Most of the Plusinae moths are nocturnal and the rhythms are driven by endogenous circadian rhythmicity. But it is also well known that the silver Y, Autographa gamma shows diurnal migration flight and Alpine species visits flowers in the daytime. We investigated the details of their rhythms using actographs. The Alpine Syngrapha nyiwonis was found to be essentially nocturnal but some, especially mated females, take flights in the daytime. Interestingly, their nocturnal activity is strictly limited in a few hours right after sunset, and they show no circadian component in the free-running state under DD or LL condition. Both of these natures seem to be adaptive to cool and unstable alpine weather. Rhythms of non- and sub-Alpine species are also discussed.

Morphometrics and insect development, genetics and evolution: integration in the *Drosophila* wing

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The techniques for shape analysis have made substantial progress since the last symposium on Insect Morphometrics at the International Congress of Entomology in Florence (1996). Morphometrics has continued to become more flexible and powerful so that specific biological questions can be addressed. The main focus in morphometrics has therefore shifted from the development of entirely new techniques to adapting and applying the existing methods to the experimental designs and protocols in use in different biological disciplines. Particularly promising areas for such new applications include genetics, developmental biology and phylogenetics. I illustrate how morphometric methods can be customised for specific biological contexts with a study of morphological integration in the *Drosophila* wing. There has long been considerable interest in the hypothesis that the anterior and posterior compartments, which contain completely separate cell lineages, are units of development, inheritance and evolution. I use different approaches to re-examine this question. Analysis of integration of fluctuating asymmetry is a means to examine covariation due to direct developmental interactions. Analyses of covariation among subsets of landmarks suggest that the anterior and posterior compartment are not any more distinct than random subdivisions of the wing. Other analyses, including analyses of mutant effects and the reconstruction of phylogenetic shape changes among related species, will be used to complete the picture.

The evolution of seasonal polyphenism in African *Bicyclus* butterflies

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Species of *Bicyclus* that live in environments with alternating dry and wet seasons show striking examples of seasonal polyphenism. We have studied the evolution of this extreme case of phenotypic plasticity using a multidisciplinary approach. The alternative seasonal phenotypes differ strongly in wing pattern, but also in life history traits. Field and laboratory experiments are used to test hypotheses about the adaptive nature of the wing patterns, whilst artificial selection provides a powerful tool for examining genetic variation and the underlying physiological and developmental bases of the polyphenism. The results will be summarized to illustrate how these different levels of biological organisation can be linked together, and to show how populations are likely to be able to modify their plasticity in response to a change in the relationship between climatic variables and the seasonal cycling in the ecological environment.

Quantitative Genetics and Geometric Morphometrics of Developmental Instability in *Drosophila subobscura*: Inbreeding, Crossbreeding and Temperature Stress using Isochromosomal Lines

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Fluctuating asymmetry (FA) is often used as a measure of underlying developmental instability (DI), motivated by the idea that morphological variance is maladaptive. Whether or not DI has evolutionary potential is a highly disputed topic, marred by methodological problems and fuzzy prejudices. Here we report the results of the effects of karyotype, homozygosity and temperature on wing form and bilateral asymmetry by using a large set of isochromosomal lines of *Drosophila subobscura* and their crosses. Our approach uses the recently developed methodologies to analyze shape configurations of landmarks within the standard statistical framework employed in studies of bilateral asymmetries, and we have extended these methods to partition the individual variation and the variation in asymmetries into genetic and environmental causal components. The analyses have revealed that both inbreeding and (sub-optimal) temperature had a substantial effect on wing form (size and shape) and wing form FA, with increasing asymmetries in inbred lines and crosses cultured at suboptimal temperature. Directional asymmetry (DA) was highly significant for wing size (left wings were consistently bigger than the right ones) and shape regardless the level of inbreeding and rearing temperature, and temperature-dependent expression of genetic variation for wing shape DA was also revealed in heterozygous crosses. No genetic variation was generally detected for wing size FA. However, the presence of genetic variation for wing shape DA introduces a bias in the estimation of FA and makes very difficult to test for genetic effects on wing shape FA. Finally, we question the suggestions that the classical linear theory cannot account for transitions between FA and DA, and that developmental processes influencing wing shape generate random differences between body sides.

Evolution of wing color in butterflies: can local adaptation initiate ecological speciation?

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The evolution of butterfly wing colouration is strongly affected by its multiple functions, such as thermoregulation, aposematic signalling and mate recognition. We investigated the genetic background of intraspecific variation in wing colour in the butterfly *Colias philodice eriphyle*, and its consequences for the role of wing colour in ecological speciation. In this species, divergent selection on wing colour across an elevational gradient in response to the thermal environment has led to increasing wing melanization at higher elevations. A breeding experiment demonstrated that wing melanization is heritable and strongly sex-linked. Also, controlled mate choice experiments in the field over a large elevational range showed an important role of wing colour in mate choice and mating success. However, a necessary condition for ecological speciation is correlated evolution of mate signals and mate preference, which may be constrained if traits have multiple functions. These results and wing colour-dependent fitness parameters obtained from several field experiments are integrated to predict the effect of multiple functions and genetic architecture on population divergence and the probability of ecological speciation.

The evolution of seasonal polyphenism in African *Bicyclus* butterflies

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Species of *Bicyclus* that live in environments with alternating dry and wet seasons show striking examples of seasonal polyphenism. We have studied the evolution of this extreme case of phenotypic plasticity using a multidisciplinary approach. The alternative seasonal phenotypes differ strongly in wing pattern, but also in life history traits. Field and laboratory experiments are used to test hypotheses about the adaptive nature of the wing patterns, whilst artificial selection provides a powerful tool for examining genetic variation and the underlying physiological and developmental bases of the polyphenism. The results will be summarized to illustrate how these different levels of biological organisation can be linked together, and to show how populations are likely to be able to modify their plasticity in response to a change in the relationship between climatic variables and the seasonal cycling in the ecological environment.

Fruit fly outbreaks in Northwestern Australia

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The Queensland fruit fly, *Bactrocera tryoni*, is Australia's worst fruit pest. Originally endemic to Queensland this species can now be found down the entire East coast where it infests most commercial crops. In contrast *B. aquilonis*, a sister species of *B. tryoni*, is found in the northwest and is not considered to be a pest. Despite this difference in pest status these two species are virtually indistinguishable by eye. It is these flies which have been implicated in recent outbreaks of fruit fly in developing horticultural regions of northwestern Australia. The outbreaks are either due to a change of behaviour in the endemic *B. aquilonis* so that they now infest commercial crops, alternatively *B. tryoni* may have been introduced into the area or there may have been a hybridisation event between the two species. We have used microsatellite analysis to firstly characterise the populations of fruit fly in the northwest and then used these profiles to identify the source of outbreak populations using assignment testing and cluster analysis. The mitotypes of outbreak flies and background populations were then used in simulations to provide a model of the outbreaks.

Assessment of bacteriophage WO involvement in *Wolbachia*-induced CI within the *Culex pipiens* complex

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The *Culex pipiens* complex include some of the most medically important mosquitoes due to their cosmopolitan distribution and their capacity to vector infectious diseases. Members of the complex differ in their ecology, behavior, physiology and vector competence. To date, no consensus has been achieved about the taxonomical status of representatives of this group. Opinions of their relationship range from distinct species with no gene flow to physiological forms with considerable introgression. Most populations of *Culex pipiens* are infected with *Wolbachia*, a maternally inherited bacterium, which causes cytoplasmic incompatibility (CI) resulting in embryonic mortality. In prior molecular characterization, we have demonstrated that *Culex* populations differ in the bacteriophage WO that is thought to infect *Wolbachia* (Sanogo and Dobson, 2004). To test the hypothesized involvement of bacteriophage WO in determining CI in *Culex*, crosses have been conducted between nine *Culex* strains that differ in their bacteriophage WO infection type. Here, we will present the crossing results and discuss the correlation between bacteriophage WO and CI type. We will also discuss the results in relation to the restriction of gene flow among naturally occurring *Culex pipiens* populations.

Intraspecific Transfer of Wolbachia In Mosquitoes By Microinjection

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Wolbachia are maternally transmitted intracellular bacteria that infect approximately 20% of insect species and over 30% of mosquito species examined. Through a mechanism known as cytoplasmic incompatibility (CI), *Wolbachia* infections can induce early embryo death when an infected male mates with uninfected female. The ability of *Wolbachia* to induce CI in its host has led to the proposal of strategies for the control of mosquitoes and the diseases that they transmit, including strategies for both the suppression and replacement of medically important mosquito populations. Both strategies require the ability to artificially generate novel *Wolbachia* infections, which will require techniques for transferring *Wolbachia* infections both interspecifically and intraspecifically (transfection). Although transfection techniques have been developed for a few other insects, there has been no successful *Wolbachia* transfection reported in mosquitoes. We have modified a transfection protocol previously used for *Drosophila*. Here we describe the development and initial tests of the modified protocol to transfer *Wolbachia* from superinfected *Aedes albopictus* (Asian tiger mosquito) into an uninfected strain of *Ae. albopictus*. Furthermore, we describe characterization of a transfected line via crosses to determine its CI phenotype. To date, the infection has been maintained in the transfected line for three generations and displays a pattern of CI consistent with *Wolbachia* transfection. We discuss the results in relation to *Wolbachia* /mosquito host interaction and its significance to applied mosquito control strategies.

Identification of source populations of invasive insects: implications for biological control, monitoring, and quarantine

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Invasive species cause severe economic and ecological damage in many regions around the world. Knowledge of an invasive species' origin benefits inspection and quarantine efforts and focuses the search for biological control agents. Cooley spruce gall adelgid (CSGA), *Adelges cooleyi*, is a native pest in the US that provides an excellent model organism for studying population genetics of invasive exotic species. The native range of CSGA resembles an inverted 'V', extending northward along the Rocky Mountains into Canada and back down the West Coast into northern California. CSGA has invaded the eastern US through an unknown number of introductions during the 20th century. Here, we report efforts to identify sources of CSGA populations introduced into the eastern US. Comparison of mitochondrial and nuclear DNA sequences among source and introduced populations revealed that all east coast introductions originated from one region within the insect's extensive native range. As expected by theory, genetic variation was decreased in introduced populations relative to populations from the insect's native range. This work demonstrates a successful application of molecular genetic tools to determine the origin of invasive species and aspects of population genetic structure, thus facilitating the development of sound, sustainable pest management strategies.

Inbreeding leads to changes in allometry between egg size and larval characters in an eriosomatine aphid (Aphididae)

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A number of studies indicate that inbreeding has deleterious effects on the viability and reproduction of organisms, but it is still unknown what mechanisms are involved in the expression of deleterious effects. Because the effects of inbreeding are expressed from the early stage of development, it is necessary to focus on hatched larvae to understand the phenotypic effects of inbreeding. Aphids are useful for studying inbreeding effects because in aphids (1) it is easy to cross males and females of the same clone (that is, selfing) and (2) hatched larvae can be used for detailed morphometric analyses. Using the aphid *Prociphilus oriens*, this study prepared eggs resulting from outbreeding and selfing and compared the allometry between egg size and morphological characters of hatched larvae between outbred and selfed groups. The allometry of larval characters against egg size varied greatly between outbred and selfed groups. Selfed larvae had significantly larger allometric coefficients for appendages than did outbred larvae, showing that inbreeding accelerated the growth of appendages. However, the growth of gonads was suppressed in selfed larvae. It is suggested that inbreeding relaxed developmental canalization on larval appendages and resulted in reduction in gonad size via a trade-off between appendages and gonads.

Rapid chill coma recovery selection in *Drosophila melanogaster*

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Resistance to low temperatures can vary markedly among invertebrate species and is directly related to their distribution. Despite the ecological importance of cold resistance this trait has rarely been studied genetically. One measure of cold resistance that varies geographically in *Drosophila melanogaster* and that is amenable to genetic analysis is chill-coma recovery. *D. melanogaster* have been selected for increased recovery time following exposure to 0°C. Selected lines responded rapidly to the selection and showed an increased resistance to a broad range of low temperatures. They also had a lower mortality following a more severe cold treatment, indicating that a general mechanism underlying cold resistance had been selected. The selection response was independent of plastic changes in cold resistance because the selected lines maintained their ability to acclimate. Life-history and stress resistance tradeoffs were also investigated showing only a tradeoff with ageing and one stress resistance trait. Selected lines were genotyped for clinally varying genes/markers and show associations with some candidate genes suggesting these genes may have a role in cold resistance.

Predicting shifts in timing of winter moth egg in response to climate change

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Climate change can lead to mistiming in phenology between different components of food chains. For instance, winter moth (*Operophtera brumata*) egg hatch has advanced more than oak (*Quercus robur*) bud burst over the past two decades. The question is whether natural selection will lead to a restoration of synchrony between winter moth and oak. Using a half-sib design, we demonstrate here that egg hatching is a heritable trait, making adaptation through selection possible. We further use a model to predict the changes in reaction norm of egg hatching at different temperatures. This approach enabled us to predict the response in the next year, under different environmental conditions, to the selection in a given year. Due to selection, the reaction norm will change in intercept but not in slope: in future eggs will need more warmth before they can hatch, regardless of what the temperature is in a given year. Because of this, timing differences between egg hatch and oak bud burst will decrease rapidly. Changes in reaction norm in response to selection thus offer a possible way of restoring synchrony between different components of a food chain, winter moth and oak.

Use of thermal data as an establishment screen for non-native biocontrol agents in the UK

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Most predatory insects and mites used in glasshouse biocontrol in the UK originate from tropical and semi-tropical climates. For this reason, the licensing system for the introduction of non-native species has operated under the assumption that winter would act as a natural barrier to the establishment of such species outside of glasshouse environments. This view has been challenged by the establishment in the wild of the predatory mite *Neoseiulus californicus* and the discovery of the predatory mirid *Macrolophus caliginosus* outside of glasshouses in winter. Whilst the impact of these species on native ecosystems is unknown, their establishment is considered undesirable. This paper describes a series of experiments used to determine a range of thermal characteristics (developmental threshold, day-degree requirement per generation, supercooling point, lethal times and temperatures, field survival) of four non-native biocontrol agents. The comparative data provide a retrospective ecophysiological explanation for the establishment of *N. californicus* and *M. caliginosus,* and also indicate that *Delphastus catalinae* and *Eretmocerus eremicus* would not survive outdoors in the UK under current climatic conditions. The experimental protocol applied to these species could be used as part of a routine screen for 'establishment potential' in the licensing system of non-native biocontrol agents in the UK.

Synchronous coadaptation in an ancient case of herbivory

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Coevolution has long been considered a major force leading to the adaptive radiation and diversification of insects and plants. A fundamental aspect of coevolution is that adaptations and counteradaptations interlace in time. A discordant origin of traits long before or after the origin of the putative coevolutionary selective pressure must be attributed to other evolutionary processes. Despite the importance of this distinction to our understanding of coevolution, the macroevolutionary tempo of innovation in plant defenses and insect counterdefenses has not been documented. Molecular clocks for a lineage of chrysomelid beetles of the genus *Blepharida,* and their Burseraceae hosts were independently calibrated. Results show that these plant's defenses and the insect's counterdefensive feeding traits evolved roughly in synchrony, providing the first macroevolutionary confirmation of synchronous plant-herbivore coadaptation. The association between these two groups of organisms was determined to be about 112 million years old, the oldest age so far for a specialized plant-herbivore association.

Genetics and evolution of photoperiodism in temperate environments

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Photoperiodism constitutes the major physiological mechanism by which arthropods adjust the seasonal programming of development and dormancy at temperate latitudes. Dispersal of the pitcher-plant mosquito, *Wyeomyia smithii*, from southern latitudes to northern Canada following recession of the Laurentide Ice Sheet 20,000 - 8,000 years ago has involved the evolution of longer critical photoperiods, thereby tracking the shorter growing seasons at increasingly more northern latitudes. Despite directional and stabilizing selection during post-glacial range expansion, the heritability of critical photoperiod increases with latitude. We attribute this increase in heritability to the release of additive from epistatic variance during sequential founder events. During rapid climate change over the last 30 years, critical photoperiod has shifted towards shorter, more southern day lengths as growing seasons have become longer. This genetic shift in photoperiodic response is detectable over a time interval as short as five years. Faster evolutionary response has occurred in northern populations where selection is stronger and genetic variation is greater. Our results suggest that other species may be in the process of analogous evolutionary responses to altered seasonality and that the composition of future biotic communities may depend on the relative abilities of their constituent species to adapt to altered seasonal interactions.

Fluctuating asymmetry of wing pattern elements and wing shape, and its relationship to migration in the butterfly *Melitaea cinxia*

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When fitness depends on morphological traits, individuals that can develop the phenotype reliably should be more fit than those, which cannot. Fluctuating asymmetry (FA; minute differences between the left and right side of a bilateral trait due to developmental errors) has therefore been used to predict the fitness of individuals in natural and sexual selection studies. In this talk we discuss how the FA of wing pattern elements and shape of the wings of both the fruitfly *Drosophila melanogaster* and the butterfly *Melitaea cinxia* can be used to obtain information on how development is stabilized and how useful FA is in a natural selection experiment. In the butterfly species a mark-release-recapture experiment was carried out to obtain information on the relationship between FA of wing pattern elements and shape, migration, past and present population sizes, dispersal, survival and mating success. The results will be discussed taking recent results into account on the mechanisms of developmental stability as obtained with *D. melanogaster*.

The effect of a resistance gene on developmental stability and fitness in a flea beetle, *Phyllotreta nemorum*

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A resistance (R-) gene enables the flea beetle *Phyllotreta nemorum* L. (Coleoptera: Chrysomelidae: Alticinae) to use the otherwise toxic *Barbarea vulgaris* ssp. *arcuata* G-type plant as a host plant in Denmark. In this study, the pleiotropic effects of the dominant R-allele at a single autosomal gene, originating from the Danish region of Kværkeby, were investigated in an isogenic background from non-resistant beetles. The presence of the R-allele causes a high mortality at the early stages of development, when the toxic G-type plants cause the high mortality in non-resistant beetles. The presence of the R-allele has little or no effect, even under suboptimal conditions, on developmental stability (measured as fluctuating asymmetry of tibialength), body size, fecundity and longevity in the adult beetles. The distribution of the beetles in the field in Denmark will be discussed and its host plant use.

Contemporary evolutionary genetics: the breadth and depth of host race evolution in bugs on two continents.

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Several species of seed bug (the soapberry bugs: Rhopalidae) from the Americas and Australia are specialized on the plant family Sapindaceae ("the 'soapberry family"), including exotic introductions and escapes. Studies on species in both Hemispheres reveal morphological, physiological, behavioral and life history adaptations to host fruits and phenologies. Many of these adaptations display rapid or contemporary evolution (in 30-50 years or 100 generations) on introduced hosts; they include traits from mouthpart morphology to wing muscle enzyme physiology. We document this evolution in soapberry bugs from Australia (*Leptocoris*) and North America (*Jadera*), showing geographic variation within continents and among host plants. The genetics underlying contemporary evolution has been analysed in a North American species using hybrids between populations adapted to different hosts as well as artificial selection. The analyses demonstrate that additive and non-additive genetic variation in the various traits contributes to contemporary evolution. Further, they reveal expected and some unexpected genetic correlations and interesting evolutionary pathways to adaptation.

A macrophysiological perspective on cold and desiccation tolerance

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Macrophysiology, or the investigation of variation in physiological traits over large geographic and temporal scales and the ecological implications of this variation, is gaining increasing attention globally for several reasons. Here, I explore large-scale variation in cold tolerance and desiccation in insects and its implications. Perhaps unsurprisingly, rates of water loss vary in an environmentally consistent fashion, such that rates are lower in dry environments. This is true of both cuticular and respiratory water loss, even though the significance of the latter and its likely response to selection have been questioned. In several species it is clear that respiratory water loss is remarkably sensitive to environmental variation and adaptive variation thereof is not inconsistent with the data. Patterns in cold tolerance are more complicated, and reflect the variety of strategies animals can adopt to survive low temperatures. Nonetheless, two major patterns are emerging from the broad scale data. First, there is decoupling of upper and lower thermal limits. Second, there are pronounced and consistent differences in the way temperate species from the Northern and Southern Hemispheres respond to low temperatures. These major patterns not only provide insights into the mechanisms underlying insect responses to cold, but also have significant implications for understanding and predicting species responses to environmental change.

Costs of salicylate- and jasmonate-mediated induced responses in *Arabidopsis thaliana*

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Fitness costs of plant defenses to herbivores and pathogens are an important aspect of the evolutionary dynamics of plant resistance. Costs of defense have been increasingly reported in the literature, yet their mechanisms remain largely unexplored. Costs may arise from increased allocation of resources to defense production at the expense of growth, decreased assimilatory capacity in highly defended plants, or in the case of induced responses, negative interactions between defense pathways that compromise resistance to certain natural enemies. I explored costs of induced responses in several *Arabidopsis thaliana* genotypes that vary in their constitutive and inducible expression of resistance traits. In the absence of benefits, highly defended genotypes, such as the constitutive expressor of systemic acquired resistance mutant, *cep1*, had lower seed production than less defended genotypes, in general. Plants induced with either salicylic acid or jasmonic acid produced less seed than uninduced plants, and costs were quantitatively associated with the induction of such defenses as trypsin inhibitors and glucosinolates, coupled with decreased assimilatory capacity. In addition, jasmonate-mediated resistance to insects was compromised in plants simultaneously induced with salicylic acid due to inhibitory defense pathway crosstalk, and an inverse relationship between bacterial and insect resistance was demonstrated for several genotypes.

Forewing shape variation in the sand cricket *Gryllus firmus*

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Populations of the sand cricket *Gryllus firmus* are composed of two wing morphs. Long winged individuals (LW) have functional hind wings, are able to fly, and thus might disperse. Short winged individuals (SW) have vestigial hind wings and cannot fly. It is commonly considered that this dimorphism is maintained by a trade-off between flight capacity and fecundity. Here, we focused on the male forewing. Since males use their forewings to produce courtship songs, this structure affects fecundity. Additionally, it is likely that some genes jointly affect the two pairs of wings. Selection for flight efficiency on the hind wings should thus indirectly affect the forewings. This implies that the fore wings might be influenced by both trading off parameters. Using geometric morphometrics we analyzed the forewing shape variations both within and among morphs. Then, using a full sib design we estimated the genetic and non genetic components of shape variation. We discuss our results in terms of selection and developmental constrains.

Molecular responses to cold and desiccation

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Heat shock proteins (Hsps) are involved in responding to both low temperature and desiccation in flesh flies, but different sets of Hsps are involved in the responses to overwintering cold hardiness, rapid cold hardening, desiccation and rehydration. The use of RNAi to knock out Hsp70 has a dramatic effect on overwintering cold hardiness, but Hsp23 appears to be much more important for rapid cold hardening. Hsp23 and Hsp70 are both upregulated by desiccation, but two other Hsps, Hsp90 and Hsc70, are upregulated when a desiccated fly is rehydrated. The rehydration response is most similar to the Hsp response to cold recovery: Hsp90 and Hsc70 are upregulted in both cases. The deployment of different subsets of Hsps thus differs from the response to heat shock, where all are upregulated simultaneously. Microarray data indicates that a number of genes, in addition to the Hsps, are involved in the fly's response to low temperature.

Phylogenetic congruence of mealybugs and their P-endosymbonts

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Tight interactions between unrelated organisms such as is seen in plant-insect, host-parasite, or host-symbiont associations may lead to speciation of the smaller partners when their hosts speciate. Totally congruent phylogenies of interacting taxa have not been observed often but a number of studies have provided evidence that various hemipteran insect taxa and their primary bacterial endosymbionts have shared phylogenetic histories. Like other hemipteroids, mealybugs (Pseudococcidae) harbor multiple intracellular bacterial symbionts that are thought to be strictly vertically inherited. Thus co-divergence of hosts and symbionts is expected. Here, robust estimates of phylogeny were generated from four fragments of three nuclear genes for mealybugs of the subfamily Pseudococcinae, and a substantial fragment of the 16S-23S rDNA of their P-endosymbionts. Phylogenetic congruence was significant, with 75% of nodes on the two trees identical. Congruence was not perfect however, and other evolutionary outcomes including host shifts could not be ruled out. It is shown that incongruence may be influenced by uncertainty in phylogenetic estimation.

Effects of short-term spontaneous mutation accumulation for life history traits in grape phylloxera

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Mutation is the source of all genetic variation but the mutation rate, effects of new mutations, and their role in the maintenance of genetic variation are still subject to controversy. These parameters are important in models of the evolution of sex and recombination, the persistence of asexual populations, and the extinction of small populations. Most estimates have come from model organisms. Here, mutation accumulation experiments were conducted with 3 clones of grape phylloxera, *Daktulosphaira vitifoliae* Fitch, a gall forming herbivore of grapes, to estimate rate of input and effects of spontaneous mutation on life history traits. This is perhaps the first such experiment using a non-model organism of economic importance. Evidence was found that mutation rates are high enough to produce heritable genetic variation over short time scales, and that the effects of mutation on these quantitative traits are deleterious on fitness. Estimates of the parameters by the Bateman-Mukai method were within the range found in previous studies. Rapid accumulation of deleterious mutations suggest that for many pest species, adaptive response under agricultural conditions may depend more on standing variation from introductions than new mutation.

Insect antifreeze proteins

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Although they have been identified in various animals, plants, fungi and bacteria, the most active antifreeze proteins (AFPs) yet discovered are found in insects. In larvae of the freeze avoiding Pyrochroid beetle *Denoides canadensis* AFPs function to prevent freezing by (1) inhibiting inoculative freezing initiated by external ice across the cuticle, and (2) by inactivating internal ice nucleators, thereby inhibiting spontaneous nucleation and promoting supercooling. Two types of insect AFPs are currently known. Those of *D. canadensis* (DAFPs) consist of 12-and 13-mer repeating units in which certain positions (especially those with threonine, serine and cysteine residues) are highly conserved. Threonines, in particular, appear to be involved in the ability of the DAFPs to hydrogen bond to ice or ice nucleators. There are tissue and temporal variations in the expression of the family of 13 variants of the DAFPs, suggesting variation in function. Optimal function of the DAFPs requires the presence of low molecular mass enhancers (glycerol, etc.) and/or certain other protein enhancers. Also, certain DAFPs have a synergistic effect on other DAFPs. In Alaska, AFPs similar to DAFPs play a role in the ability of larvae of the Cucujid beetle *Cucujus clavipes* to supercool to extreme temperatures, in some cases below -80C.

Canalization and cryptic variation in *Drosophila melanogaster*.

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Canalization has been discussed a great deal with respect to evolutionary developmental biology, however there is a lack of consensus as to a precise definition of canalization itself. This has resulted in a large number of studies exploring different aspects of the same question with no common reference. I will discuss some methodological and statistical approaches to addressing questions of canalization, and cryptic genetic variation, and provide some empirical examples of how these can help to improve our understanding of canalization. Furthermore, I will summarize the results of a recent association mapping study designed to explore the genetic basis of cryptic variation.

Evolutionary and functional genomics of wing shape in *Drosophila melanogaster*.

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While most organisms vary considerably in aspects of shape, surprisingly little is known about the genetic basis of shape variation. Using the wing of *Drosophila* as a model system, I will present the results of a functional genomics study of wing shape, and how mutants in known wing patterning genes affect shape. Furthermore I will discuss the results of association mapping studies between natural polymorphisms in two genes, *Epidermal growth factor receptor* and *hedgehog*, with wing shape.

Diversity in the weapons of sexual selection: horn evolution in the beetle genus *Onthophagus*.

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Both ornaments and weapons of sexual selection frequently exhibit prolific interspecific diversity of form. Yet, most studies of this diversity have focused on ornaments involved with female mate choice, rather than on the weapons of male competition. With few exceptions, the mechanisms of divergence in weapon morphology remain largely unexplored. Here, I examine the evolutionary radiation of one type of weapon: beetle horns. I use partial sequences from one nuclear and three mitochondrial genes to develop a phylogenetic hypothesis for a worldwide sample of 39 species from the dung beetle genus *Onthophagus* (Coleoptera: Scarabaeidae), and then use these data to trace the evolutionary radiation of horns. Although this limited sample of one of the world's most species-rich genera almost certainly underestimates the number of evolutionary events, it reveals prolific evolutionary lability of these exaggerated sexually selected weapons (over 24 separate gains and losses of five different horn types).

Genotype to phenotype: physiological control of trait size and scaling in insects.

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For almost a century, biologists have used trait scaling relationships (bi-variate scatter-plots of trait size versus body size) to characterize phenotypic variation within populations, and to compare animal shape across populations or species. Scaling relationships are a popular metric because they have long been thought to reflect underlying patterns of trait growth and development. However, the physiological mechanisms generating animal scaling are not well understood, and it is not yet clear how scaling relationships evolve. Here we review recent advances in developmental biology, genetics, and physiology as they pertain to the control of growth of adult body parts in insects. We summarize four mechanisms known to influence either the rate or the duration of cell proliferation within developing structures, and suggest how mutations in these mechanisms could affect the relative sizes of adult body parts. By reviewing what is known about these four processes, and illustrating how they may contribute to patterns of trait scaling, we reveal genetic mechanisms likely to be involved in the evolution of insect form.

Impact of 1997-1998 ENSO-induced fires and logging on the genetic diversity of *Upadia theda* Felder (Lycaenidae) in Borneo.

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Drastic changes in population size as a result of disturbance can severely affect genetic variation. However, although severe disturbance is assumed to reduce levels of diversity, intermediate levels of disturbance, on the contrary, can lead to elevated levels of diversity. Here, we investigated the temporal variation in genetic diversity in populations of the butterfly species *Upadia theda* Felder (Lycaenidae), that were differentially affected by disturbance in East Kalimantan, Indonesia. We focused on the two main forms of disturbance that affect tropical rain forests, namely logging and ENSO-induced drought and burning. The populations were sampled over a four-year period in pristine, logged and burned areas before and after 1997-1998 ENSO-induced event. We assessed genetic variation within and between samples by analysing mitochondrial DNA sequences (mtDNA control region+ 5'end of 12S) and five microsatellite loci. Both molecular markers gave concordant results with respect to the temporal pattern of genetic diversity within disturbed areas: a significant loss of genetic diversity was observed in burned areas compared to pristine areas, but there was an increase of observed genetic diversity within logged areas. Gene flow between *Upadia theda* populations appeared, furthermore, to be highly restricted at the geographic scales assessed (less than 100 km). Importantly, our genetic diversity results are in line with previous published studies on temporal and spatial patterns of butterfly species richness.

Human-induced changes in organism distributions and the evolution of insect diets

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Human activities have caused many species to expand their ranges and humans have introduced organisms into many new and often very novel environments. These human-induced changes in organism distributions create natural experiments with which we can study ecological and evolutionary processes that otherwise could only be inferred indirectly. I examine the consequences recent changes in plant distributions for the evolution of insect diet. Using a model system of diet evolution is seed beetles (Coleoptera: Chrysomelidae: Bruchinae) I examine evolutionary constraints on adaptation, the role of life history plasticity, and the importance community composition in mediating diet expansion and adaptation.

Investigating the genetic basis of phenotypic variation in *Drosophila* egg shell patterning.

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Patterning of the insect egg shell provides an excellent system for exploring the molecular basis of phenotypic variation. In *Drosophila melanogaster*, two dorsal-anterior respiratory appendages are produced in response to signaling through the *Epidermal growth factor receptor* (*Egfr*). Previous empirical and theoretical work implicates *Egfr* function in both intra-specific variation for dorsal appendage spacing on the eggshell, as well as inter-specific differences in dorsal appendage number and location. To test the hypothesis that genetic variation in *Egfr* contributes to variation in egg shell patterning, we have made use of naturally occurring intra-specific variation for dorsal appendage spacing as a model quantitative trait. An initial association study suggests that two common polymorphisms in *Egfr* are involved in trait expression. We will report on our efforts to confirm the involvement of these SNPs as well as efforts to identify additional loci that contribute to dorsal appendage spacing.

Microbial diversity in termite guts and their role in mineral transformation of soil

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Termite a macro invertebrate act as soil engineers in tropical ecosystem modifying the environment in both biotic and abiotic components. Termites have a major effect on the physical properties, increases water holding capacity, control soil erosion and also improves the crop sustainability. Polysaccharide producing bacteria *viz., Azotobacter* and *Beijerinckia* isolated from the foregut of termites which are responsible to fix atmospheric nitrogen. Soil aggregation improves the physical parameters - porosity (40.3%), bulk density (1.33 g/cc), particle density and water holding capacity (39.8%), chemical parameters – increased organic carbon (0.098%) and available nitrogen (70 ppm). Incorporation of *Beijerinckia* sp. in acid soils provides additional attributes for crop production and environmental sustainability. The fungi *viz., Termitomyces* isolated from the fungal garden involved in the degradation of leaf litter in terrestrial ecosystem and also used as edible mushroom due to its medicinal value. Hence termites, are not only pests, but also play an important role in sustaining soil ecosystems.

Costs and benefits of glandular trichomes in *Datura wrightii*: why be Sticky?

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The relationship between costs and benefits of herbivore resistance predicts the evolution of resistance. *Datura wrightii* (Solanceae), is dimorphic for leaf trichome type. Some plants are covered with short, non-glandular trichomes ("velvety plants"), while others are covered by longer, glandular trichomes that secrete acyl sugars ("sticky plants"). Glandular trichomes confer resistance to many insects but not all. In the absence of herbivores, glandular trichomes are costly because sticky plants produce 45% fewer seeds than velvety plants. Sticky plants grow larger than velvety plants, and this may confer a competitive advantage. In a three-year field study, plant competition reduced plant seed production by 76 – 87% overall. Competition was asymmetrical and favored sticky plants because sticky plants produced 42% more seeds when competing with velvety plants than when competing with other sticky plants. Conversely, velvety plants produced 53% fewer seeds when competing with sticky plants than when competing with other velvety plants. Increased competitive ability is an additional potential benefit of the suite of traits associated with the production of glandular trichomes in *D. wrightii* and may contribute to the maintenance of the trichome dimorphism in natural *D. wrightii* populations.

The mating acceptance rate of female was higher for large males than for small males.
Males fought with other males over seeds using his fore legs, and the larger males won in all cases.
Females were attracted to some odour of male, and it was more effective when males were accompanied with seeds.

In this species, copulation interval is much longer in females, which makes OSR male-biased and competition over females severe. We conclude that males adopt Resource Defence Mating System, in which they defend seeds and/or area from other males, and attract females with some odour. Therefore, sexual selection favours larger male body size.

Wing shape, wing size and fitness in *Trichogramma* wasps

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Trichogramma wasps are tiny egg parasitoids widely released commercially to control moth pests. To evaluate the impact of wing shape and wing size on wasp fitness, we released wasps at a central point and compared the morphology of female wasps in a release sample with females that successfully located eggs of target hosts. Three species-host-plant combinations were evaluated: *T. carverae* attacking lightbrown apple moth (*Epiphyas postvittana*) on grapevines, *T.* nr. *brassicae* attacking *Helicoverpa* spp. on tomatoes, and *T. brassicae* attacking European corn borer (*Ostrinia nubilalis*) on corn. In each case, wing size was calculated as centroid size and wing shape was assessed using Procrustes superimposition. Wing size of wasps had an impact on field fitness in all cases, but in *T. brassicae* this association was complicated by the fact that wing size effects changed with distance from the release point and time since release. Wing shape had no impact on field fitness in *T. brassicae*, but did influence field fitness in *T. carverae* and *T.* nr. *brassicae*. Changes in wing shape associated with high fitness were different in the two situations. Rearing host influenced wing shape. In all cases shape effects appeared to be at least partly independent of allometry.

Clinal variation and the genetics of stress resistance

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Clinal variation for traits within species provide an outstanding resource to identify traits and genes under selection due to climatic factors. The most well known clines in insects are for body size, but clinal variation in a range of other traits has also been identified. One of the most intensively studied clines is the *Drosophila melanogaster* cline along the east coast of Australia where populations can be collected along a transect ranging from temperate Tasmania to tropical Far North Queensland. There are opposing patterns along this transect for resistance to cold and heat stress, as well as clinal variation for evasion of stressful winter conditions by altered patterns of egg production. We are now dissecting these patterns phenotypically and genetically to identify the physiological and genetic basis of adaptive shifts in these traits. QTL mapping following crosses between populations from cline ends indicate regions of the genome associated with traits, while QTL mapping and association studies from mid-latitude populations provide evidence for specific candidate locus-trait associations. We show how the performance of selected lines in field cages can validate traits under selection and how parallel clines in related species can validate the adaptive nature of the clines.

Cryoprotective dehydration – cold tolerance in permeable invertebrates

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Besides the two well-known cold hardiness strategies, freeze avoidance and freeze tolerance, a third, and fundamentally different, mechanism to survive freezing temperatures by dehydration exists in organisms with highly permeable integument. When soil water in their environment freezes, they dehydrate due to the difference in water vapour pressure between the supercooled body fluids and the surrounding ice. Dehydration continues until vapour pressure equilibrium between internal fluids and the surrounding ice is reached. Recent studies have shown that this cold hardiness mechanism is widespread in soil invertebrates (e.g. Collembola) and coincides with high cuticular permeability to water. In such animals, supercooling capacity is of little or no importance for winter survival. Dehydration triggers physiological changes in Collembola, such as accumulation of sugars and polyols, expression of heat shock proteins, and shifts in the phospholipid composition of membranes. These changes are likely to promote cold tolerance in a dehydrated state.

Parasitoid foraging behavior in a life history context

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In the 1980's a synthesis between Behavioral Ecology and Life History Theory was initiated by including life history parameters into state dependent optimal foraging models. To test these models, we collected empirical data showing that in the Australian parasitoid *Asobara persimilis* (Hymenoptera: Braconidae), genetic differences in life history traits correlate with differences in foraging behavior. *A. persimilis* occurs along the entire Australian east coast. We studied life span, patch time allocation and oviposition behavior of parasitoid populations from the Adelaide, Melbourne, Sydney and Brisbane regions. Life span increased along a gradient from Adelaide to Brisbane. Correlated to this, patch residence times and oviposition activity decreased along the same gradient. These results are in accordance with foraging models predicting a negative correlation between life expectancy of time-limited foragers and the degree to which they should exploit patches of a resource with a clumped distribution.

Exploring winter energy consumption as a limitation to the southern range limit of the goldenrod gall fly (*Eurosta solidaginis*) in late goldenrod (*Solidago gigantea*)

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The goldenrod gall fly, *Eurosta solidaginis*, is a gall-forming insect that has diverged into two host races on its hosts *Solidago gigantea* and *S. altissima.* These host races show appreciable reproductive isolation and genetic differentiation. Both host plants range from Ontario to Florida in eastern North America. Flies of the *altissima* host race can be found virtually everywhere its host plant is present, but *gigantea* flies are only present in the northern tier of its host's range. We performed a series of experiments to identify factors restricting the *gigantea* host race to northern locales. Correlation analysis suggests that the southern limit of *gigantea* flies is constrained by warm winters, likely because warmth increases energy consumption. *Gigantea* flies are smaller and less fecund because *S. gigantea* senesces earlier (thus ceasing feeding earlier in the autumn) than does *S. altissima.* Being smaller, the *gigantea* host race has less energy to support metabolism during winter and egg production the following spring. We will present our ongoing efforts to produce a model of winter energy use for goldenrod gall flies across their range, and consequent range limitations. Our work will stand as a model for predicting shifts in geographic distributions with global climate change.

Sources of variation in costs of plant resistance to herbivores

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A meta-analysis of 70 studies assessing the relationship between plant resistance and fitness has been conducted to examine the importance of several sources of variation in the fitness cots of resistance. The greatest costs were observed under uncontrolled environmental conditions in the field, at high levels of nutrient availability, and for defenses associated with alkaloids and phenolics. The prevalence of costs in the field and their persistence in the presence of herbivores both imply that fitness costs may arise not only due to trade-offs in the allocation of common limited resources between defense, growth and reproduction within an individual plant (direct allocation costs), but also through interactions between plants and external factors in their environment (ecological and opportunity costs). Cumulative meta-analysis revealed the evidence of publication and citation bias in studies on costs of plant resistance. The early studies conducted in 1975-1990 reported significant negative correlations between defense and fitness measures. However, after publication of several theoretical studies that have provided theoretical justification for the absence of costs, the magnitude of the reported costs decreased and became non-significant in the early 1990s. Furthermore, studies reporting significant costs of resistance received more citations than those, which reported no evidence of costs.

The evolution of egg retention and sex allocation: a study of facultatively ovoviviparous thrips

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Idolothripine thrips provide an unparalleled opportunity for investigating the evolution of reproduction. In many species, females are facultatively ovoviviparous/viviparous, and individuals choose among egg laying, live birth or both. As thrips are haplodiploid, females have some control over the sex of their offspring. We present our preliminary phylogeny and contrast work for the subfamily, and a case study of the Japanese idolothripine, *Bactrothrips brevitubus*. In this species, both sexes are produced by oviparity but only males are produced by ovoviviparity, so that sex allocation and reproductive mode act in tandem. Local resource competition between females for food and egg-laying sites may select for ovoviviparity, as females can retain their eggs until a better rearing site is located. Predatory mites could also select for ovoviviparity, as they consume the eggs but are non-fatal to larvae and adults. Population-wide sex ratios are generally unbiased, but our DNA and life history data indicate that the sex ratios are not Fisherian and are split according to the dominant reproductive mode in a patch. We consider how females tradeoff the best reproductive mode and sex allocation strategies. More broadly, we consider how egg retention and sex allocation may have coevolved in the subfamily.

Rapid cold-hardening: mechanisms and ecological significance

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More than 15 years ago we reported a rapid cold-hardening (RCH) response that protects various species of insects and related taxa against cold shock (non-freezing) injury. This swift acclimatory response to a brief exposure at a moderately low temperature allows the insect to survive a subsequent cold shock that would otherwise be lethal. Although little is known about the physiological mechanisms underpinning this response, recent evidence suggests that modifications of membrane fluidity and stress proteins play a role. RCH can also occur in isolated cells directly exposed to low temperature. Originally, we hypothesized that the RCH response allowed insects to enhance "instantaneously" their cold tolerance in a thermally variable environment. Recently, we obtained strong support for this hypothesis by demonstrating that RCH occurs under ecologically relevant cooling rates and diurnal thermoperiods. Furthermore, the rapid acclimatory response is not restricted to cryoprotection, but also functions to enhance organismal function at multiple levels, including courtship and mating, in response to environmental cooling at moderate to high temperatures. Thus, it is clear that RCH has broad ecological significance because it allows insects to continuously fine-tune organismal response to match even modest changes in environmental temperature that occur throughout the year.

Costs and benefits of resistance and tolerance to herbivores: the importance of gene x environment effects

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Trembling aspen (*Populus tremuloides*) is the most widespread tree species in North America and the most genetically variable plant species known to science. Extraordinary genetic variation of this clonal species extends to traits such as rates of growth and levels of chemical (salicylates and tannins) defense. Given that aspen is routinely subject to expansive defoliation events (e.g., large aspen tortrix, forest tent caterpillar) the question arises as to why defense polymorphisms persist in natural populations, i.e., why do not all genotypes express maximum levels of defense? The emerging answer involves both environmental and ontogenetic influences on the expression of chemical defense. For young aspen trees, high commitment to chemical defense incurs a significant cost in terms of growth in resource-poor, but not resource-rich, environments. Thus, in the absence of herbivory, highly defended trees are disadvantaged because slow growth may limit competitive ability. As trees mature, salicylate-based defenses tend to decline, extensive root systems develop, and trees appear to shift their defensive strategy to tolerance. Tolerance itself is also genetically variable and influenced by resource availability.

Sexual isolation in North American fruit flies: reinforcement, reproductive character displacement and the role of courtship song

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Differences in the courtship song of *Drosophila* males, produced by wing vibrations, have been implicated in sexual isolation between several species and races through female discrimination. The range of *Drosophila persimilis,* California, Oregon and Washington State, is completely contained within that of its sibling species *D. pseudoobscura* which extends as far east as Texas. The species are morphologically identical but sexually isolated. Although male courtship appears to be indiscriminate, females of these species discriminate against mating with heterospecific males. Females from areas where they co-occur discriminate more than those collected from allopatric locations. This has been suggested to be an example of speciation by reinforcement. Despite identifying species differences in both major song parameters, interpulse interval (IPI) and intrapulse frequency, previous work has suggested that these song differences are not responsible for sexual isolation. In this study playback experiments of synthetic *D. pseudoobscura* and *D. persimilis* song demonstrate clearly that females do indeed discriminate between conspecific and heterospecific courtship song. Reinforcement may be as a result of a narrowing in female preferences in sympatry or of reproductive character displacement (RCD) in a male courtship trait. The courtship songs of *D. pseudoobscura* from both sympatric and allopatric locations are examined for evidence of RCD.

Adaptive responses by Australian burrowing cockroaches to fluctuating Neogene and Pleistocene environmental conditions: evolution in parallel and reverse?

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Over the last ~15 million years, many areas in Australia once covered by rainforest have been transformed into zones of savanna, shrubland and sclerophyll forest, primarily due to long periods of arid climatic conditions. Understanding how the ancient ancestors of Australia's unique extant fauna responded to these changes is of singular interest. We have recently provided phylogenetic evidence that Australian-native soil-burrowing cockroaches evolved from wood-feeding ancestors following an invasion of the latter from Asia into Australia. The transition from wood to soil burrowing was most likely a result of aridification, which would have led to the disappearance in many areas of wood suitable for burrowing. We have extended our study to include all known soil-burrowing cockroach species, as well as most Australian burrowing cockroach species. Phylogenetic analyses based on mitochondrial COII data suggest that the soil-burrowing habit may have evolved on more than one occasion. An alternative explanation is that it evolved only once, but has been followed by 're-evolution' of the wood-burrowing habit. Approximate times of divergence between key taxa will be estimated using molecular dating methods, allowing a preliminary examination of whether these transitions occurred at similar or different times.

Does predation maintain plasticity in *Bicyclus anynana*?

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The butterfly *Bicyclus anynana* exhibits phenotypic plasticity involving the wet season phenotype which possesses marginal eyespots on the ventral surface of the wings and the dry season form that lacks these eyespots. We examined the adaptive value of phenotypic plasticity of *B. anynana* in relation to the defence mechanisms of crypsis and deflection. We assessed the visibility differences between spotless and spotted butterflies against backgrounds of brown (y season) or green (wet season) leaves. Spotless butterflies were highly cryptic and less predated by adult bird predators than spotted ones when presented against brown leaf litter. The advantage of crypsis disappeared in the wet season habitat as both forms were equally visible. In later experiments, naïve birds presented with resting butterflies in the wet season habitat tended to learn more rapidly to capture spotless butterflies, suggesting a slight selective advantage of possessing eyespots. Marginal eyespots increased significantly the escape probability of butterflies that were attacked by naïve birds compared to adult birds, although there were no differences in prey capture success within naïve predators. Our results show that natural selection acts against eyespots in the dry season favouring crypsis whereas in the wet season it may favour eyespots as deflective patterns.

The peppered moth: decline of a Darwinian disciple

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The evolution of melanism in the peppered moth, following the industrial revolution in Britain, is the most cited example of an observed heritable change that resulted from Darwinian evolution. The reason for this is that the change was visually obvious, was observed, and could be explained by Darwin's central mechanism of evolution - natural selection. Further, the reason that the black form was fitter than the pale form was easy to understand: against lichen denuded and smoke blackened bark, the black moths were better camouflaged, and so were less often found and eaten by birds. Recently creationists have seized upon design flaws in Bernard Kettlewell's classical experiments in the 1950s. They claim that these flaws, and more recent allegations of fraud aimed at Kettlewell and a conspiracy of silence among evolutionary biologists, particularly in Britain, fatally undermine this cornerstone of Darwinian evolution, and therefore the basic theory of evolution by natural selection. Many schools in the U.S.A. have dropped the peppered moth case from biology teaching, and pressure is growing elsewhere. Most biologists who have worked on the peppered moth take the view that although Kettlewell's experiments did contain design flaws, these flaws would only have affected the quantitative accuracy of his tests; not the qualitative interpretation that differential bird predation has played a major role in the evolution of melanism in the peppered moth. The question I will address is whether melanism in the peppered moth should still be regarded as an example, par excellance, of Darwinian evolution in action.

Origin and evolution of seasonal adaptations: ground crickets in tropical, subtropical and temperate climatic conditions

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The ground cricket species complex, *Dianemobius nigrofasciatus* and *D. fascipes*, is probably of tropical origin and widely distributed from tropical to temperate regions in East Asia. It might be surmised that as peripheral populations dispersed northward, they encountered gradual changes in climatic conditions, evolved locally different types of seasonal life cycle, and egg diapause became the core of adaptation to overwintering in the northern climate. I pursued the origin of egg diapause and the associated patterns of nymphal development in local populations at low (tropical and subtropical) latitudes and found dual responses (egg diapause and delayed nymphal development) to short days in some of them. In other tropical populations a few eggs showed delayed hatching. The occurrence of such eggs might represent 'life-cycle polymorphism' to cope with unpredictable risks after hatching and might be the source of evolution for egg diapause and overwintering in the north.

Ontogenetics of QTL: the genetic architecture of trichome density over time in *Arabidopsis thaliana*

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Although much is known about the molecular genetic basis of trichome development in *Arabidopsis thaliana*, less is known about the underlying genetic basis of continuous variation in a trait known to be of adaptive importance: trichome density. The density of leaf trichomes is known to be a major determinant of herbivore damage in natural populations of *A. thaliana* and herbivores are a significant selective force on genetic variation for trichome density. A number of developmental changes occur during ontogeny in *A. thaliana*, including changes in trichome density. I used multiple interval mapping (MIM) analysis to identify QTL responsible for trichome density on both juvenile leaves and adult leaves in replicate, independent trials and asked whether those QTL changed with ontogeny. In both juvenile and adult leaves, I detected a single major QTL on chromosome 2 that explained much of the genetic variance. Although additional QTL were detected, there were no consistent differences in the genetic architecture of trichome density measured on juvenile and adult leaves. The finding of a single QTL of major effect for a trait of known adaptive importance suggests that genes of major effect may play an important role in adaptation.

Regulation of a plant allelochemical-inducible detoxification gene via the aryl hydrocarbon response cascade

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The promoter of *CYP6B1*, a cytochrome P450 monooxygenase (P450s) that metabolizes furanocoumarins (i.e. xanthotoxin) found in the specialized diet of the swallowtail caterpillar *Papilio polyxenes*, utilizes several elements for basal and xanthotoxin-inducible expression, including the xenobiotic response elements to xanthotoxin (XRE-Xan) and aryl hydrocarbons (XRE-AhR). In mammalian systems, XRE-AhR mediates induction of *CYP1A1* by aryl hydrocarbons via the aryl hydrocarbon receptor (AhR) and its heterodimeric partner, ARNT. To determine if AhR-like and ARNT-like proteins from *Drosophila melanogaster*, spineless-aristapedia (Ss) and tango (Tgo), are capable of activating *CYP6B1* transcription via its XRE-AhR element, Ss and Tgo proteins were coexpressed with the wildtype *CYP6B1* promoter:CAT construct in an Sf9 cell transfection system. These studies indicate that basal and xanthotoxin-inducible expression are significantly increased in the presence of Ss and that xanthotoxin-inducible expression is increased in the presence of Tgo. Coexpression of Ss/Tgo with the *CYP6B1* promoter constructs containing mutations in the XRE-Xan or XRE-AhR elements indicate that mutations in the XRE-AhR element reduce basal and xanthotoxin inducible expression but retain low levels of activation in the presence of Ss/Tgo. These results suggest that the *CYP6B1* promoter employs conserved elements of the aryl hydrocarbon regulatory cascade for basal and xanthotoxin-inducible expression that are shared between vertebrates and invertebrates (XRE-AhR).

"*Drosophila melanogaster* gene expression in response to a virulent endosymbiont "

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Wolbachia pipientis is an obligate intracellular bacterium present in a wide range of insect species. While the bacterium is known for its ability to manipulate host reproductive biology, most strains cause little direct harm to host tissues. One exception is a strain called "popcorn" that infects *Drosophila melanogaster*. As the fly ages the bacterium replicates to large numbers, leading to rupture of host cells and shortened insect lifespan. This over-replication is aberrant in comparison with other *Wolbachia* infections whose densities remain stable or decline with insect age. Previous work indicates that *Wolbachia* infections do not induce or actively suppress an innate immune response in *Drosophila melanogaster*. Because the popcorn infection leads to cell rupture we predicted that this strain would likely induce host defenses. Using a microarray approach we have examined the effect of popcorn infection on host gene expression as the host ages. Overall, gene expression decreases with age in response to popcorn infection. The bacterial specific response resulting in antimicrobial peptide production is not activated. The most common class of upregulated genes, however, were serine proteases; including elastases and trypsins. Serpins, the regulators of these proteases are also highly expressed, as are other genes encoding general defense response proteins.

Evolutionary dynamics of *w*Au-like *Wolbachia* strains in Neotropical *Drosophila* species

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We have screened 29 American strains of the Neotropical species *Drosophila willistoni* for the infection status with the -proteobacterium *Wolbachia wsp* PCR diagnostics and Southerns blots. Although *Wolbachia* is absent in older strains of *D. willistoni* collected before the 1970s more recent samples derived from natural populations all over the American continent are universally infected with the A-group *Wolbachia* strain named *w*Wil. *w*Wil of *D. willistoni* is identical to *w*Au of *D. simulans*. Like *w*Au in *D. simulans w*Wil is not inducing cytoplasmatic incompatibility (CI) in *D. willistoni*. In contrast to *w*Au a strain infecting both germline and somatic tissues *w*Wil is exclusively targeting the primordial germline cells of *D. willistoni* embryos. This tissue tropism of *w*Wil and its temporal and spatial biogeographically pattern strongly support (*i*) a long-term evolutionary relationship between *w*Wil and its host species *D. willistoni* in some isolates; (*ii*) its modern day infective spreading throughout *D. willistoni* American populations, and (*iii*) our assumption that this Neotropical *Drosophila* species can be regarded as the most likely donor source for a recent *Wolbachia* infection of immigrating *D. simulans* with the strain *w*Au. Furthermore new *Wolbachia* strains derived from Neotropical *saltans* group species do harbor polymorphic *w*Au-like *Wolbachias*.

Mechanism and evolution of caste polyphenism in social insects

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Phenotypes of organisms are not determined completely genetically, but vary according to environmental factors (phenotypic plasticity). Some organisms express several discrete adaptive phenotypes (polyphenism). Social insects possess a few types of individuals (castes) in their colonies, to which tasks are allocated. Here, I introduce our case studies related to caste polyphenism in ants and termites. We focus on the developmental mechanisms of cast-specific characters, such as alate wings or soldier mandibles. In ants, the developmental fate of caste is determined in the early stage of postembryonic development, but apoptotic degeneration occurs in the wing primordia of future workers. As apoptotic wing degeneration was observed in two phylogenetically distant groups of ants, this phenomenon is suggested to be common in many ant species. On the other hand, all termite species possess distinct sterile soldiers, which have specific morphology suitable for defense. We isolated genes related to soldier differentiation and analyzed the expression profiles of those genes, in order to understand the mechanism of caste differentiation and the link between molecular and social evolution. I will compare these studies, in terms of the alteration of body plan in response to environmental signals, and discuss the evolutionary process of the interaction between ontogeny and environment.

Possible reproductive isolation as an indirect consequence of adaptation to different environments: pleiotropic effect of clock genes

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Clock genes pleiotropically controlling development time, circadian rhythm and time of mating can cause an allochronic reproductive isolation in a model insect, the melon fly *Bactrocera cucurbitae*, which mate once a day at dusk. A series of experiments including artificial selection and behavioural analyses showed that flies with shorter developmental and circadian periods (ca 22 h of locomotor activity rhythm) mated 5 h earlier in the day than those with longer periods (ca 30 h). Mate choice tests demonstrated significant pre-mating isolation between populations with short and long circadian periods. Pre-mating isolation did not occur when the mating time was synchronized between the two populations by photoperiodic controls, indicating that reproductive isolation is due to variations in the time of mating and not any unidentified ethological difference between the two populations. The fluctuation of *per* gene mRNA expression under DD is closely correlated to the locomotor rhythm in each population with short and long periods in *B. cucurbitae.* The results suggest that a clock gene might be involved in the allochronic pre-mating isolation. I discuss allochronic reproductive isolations as an indirect consequence of adaptation to different environments in view of a pleiotropic effect of clock gene controlling time-related traits.

The development and evolution of novel phenotypes: a case study on beetle horns

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How do ecological, developmental, and genetic mechanisms interact in the origin and diversification of novel phenotypes? This question is at the center of my research on the evolution and development of beetle horns, a morphological novelty that upon invention has undergone dramatic diversification on a variety of levels. Horn expression is modulated in the context of alternative male morphologies, sexual dimorphisms, between-population differences in allometric scaling, and between-species differences in the number and location of horn development. Combining morphometric, candidate-gene, and ecological approaches my research aims to integrate a genetic and developmental understanding of phenotype expression into the behavioral and ecological contexts within which different phenotypes function. In my talk I will first focus on how a developmental genetic understanding of horn expression can inform us about the origins of non-linear scaling relationships, sexual dimorphisms, and the parallel gain and loss of horns in similar and different body regions. I will then explore which ecological and social conditions contribute to and guide evolutionary changes in beetle horn development. Beetle horns and horned beetles emerge as an outstanding model system for studying the interplay between ecological and developmental mechanisms is the origins of phenotypic diversity.

Attempts to search for host genes affected by intracellular symbiotes in their expression level: transcriptome analyses with silkworm cDNA microarray

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Arthropods harbor intracellular bacteria, such as *Wolbachia* and *Cardinium* (CFB [*Cytophaga-Flavobacterium-Bacteroides* phylum] bacteria). These facultative bacteria cause reproductive alteration in their hosts such as thelytokous parthenogenesis, male killing, feminization and cytoplasmic incompatibility. As a part of the attempt to investigate the effects of these bacteria on hosts, we examined gene expression in bacteria-infected and non-infected cells. *Wolbachia* of the small brown planthopper *Laodelphax striatellus* and Is-CFB (CFB bacteria isolated from *Ixodes scapularis*) were cultivated in silkworm cell lines (BmN4 and NIAS-Bm-aff3). A cDNA microarray, which contains about 6,000 cDNA clones of the silkworm, was used to identify the genes that were affected in the expression level by the infection of *Wolbachia* or Is-CFB. No clearly up-regulated genes were found by *Wolbachia* infection, but one transcript was down-regulated. Is-CFB infection raised expression level of a few genes.

Female genitalia and sexual selection: a morphological study of cockroach genitalia

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The male genitalia of Hexapoda are often complex and species-specific in form. Several hypotheses have been proposed to explain this. This study tests three hypotheses; Lock-and-Key, Female Choice and Male-Female Conflict by studying the genitalia of four species of cockroaches. The interacting parts of cockroach genitalia are on the morphological outer side of the body and are partly sclerotised in both sexes. Furthermore, cockroach male genitalia are highly asymmetric and the direction of asymmetry has been reversed several times. These traits make cockroach genitalia easily accessible, morphologically variable, and thus highly suitable for the study of genitalic evolution.
Contrary to "Lock-and-Key," the asymmetry of the male genitalia is not matched by asymmetry in the female genitalia. As predicted by "Female Choice", mechanoreceptors are found in the areas of genitalic interaction, but a reversal of male genitalia asymmetry does not influence their placement. There is quite strong evidence against "Male-Female Conflict" as the female genitalia facilitate gripping by the male through a ledge or other holding points. However, in water-striders "Male-Female Conflict" is well documented. This suggests that none of the proposed hypotheses can explain genitalic evolution in the Hexapoda by itself, thus the ultimate explanation must be something different.

Genetic diversity and oviposition host preference of *Bemisia tabaci* (Gennadius) (Homoptera: Aleyrodidae) associated with four crops in Ghana

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The genetic and behavioural diversity of populations of the polyphagous virus vector, *B. tabaci* infesting cassava, tomato, garden egg and okra in Ghana were assessed. Seventy-six individuals collected from four hosts in three locations were screened by RAPD-PCR, using five primers (OPA 02, OPB 08, OPC 05, OPD 16 and OPI 16). Cluster analysis using the Dice coefficient revealed two genetic clusters at the 45% level of similarity associated with cassava and okra. Crop association contributed more to the observed variability than geographical isolation. Two-choice oviposition preference bioassay was done in a petri-dish cage. Cassava whiteflies significantly ($p < 0.01$) preferred cassava to any other host, but preferred garden-egg or tomato in the absence of cassava. Tomato populations significantly ($p < 0.05$) preferred tomato and okra, while okra whiteflies did not oviposit on cassava. These results corroborate the existence of two distinct biotypes: the cassava biotype and the polyphagous biotype. Tomato, garden egg and eggplant are potential common hosts for the two and may be reservoir hosts for under intensive systems or in rotation programmes. It is suggested that whitefly management on the main hosts should accommodate the possible spill of resistance from these hosts into cassava and vegetable crops.

Molecular characterization of different biological forms of *Anopheles stephensi* **Liston in Iran**

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Malaria is still one of the main health problems in south and southeast provinces of Iran. Mosquitoes of *Anopheles stephensi* are the most important malaria vector in these regions. Based on egg ridges three biological forms of type, intermediate, and *mysorensis* has been reported within this species which shows different vector capacity and competence. Here we study the sequence variation of 1500 bp of mtDNA cytochrome oxidase subunit I (COI) of these biological forms to find molecular marker for diagnostic purposes. Different biological forms were collected from different parts of the country and were subjected to PCR-RFLP or PCR sequencing assays. Results of PCR-RFLP showed no polymorphism between different biological forms or populations. This was confirmed by comparing 1400 bp sequences of COI and COII regions which showed a high similarity between forms. However there were two haplotypes between populations of *mysorensis* form. These findings are in agreement with previous hybridization, cytological, and molecular studies stating a great gene flow between populations of *An. stephensi.*

mtDNA PCR-RFLP and PCR sequencing revealed significant genetic variation among *Anopheles superpictus* populations in Baluchistan, Iran

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Mosquitoes of *Anopheles superpictus* are one of the main malaria vectors in Iran and present in most parts of the country. This taxa play different rules in malaria transmission in different areas of its geographical distribution. This study has been conducted to investigate rate of genetic variation between different populations of *An. Superpictus* in Baluchistan, the most important malarious region in Iran. A portion of about 1512 bp long of mitochondrial DNA cytochrome oxidase subunits I and II were amplified and were assayed by either RFLP or direct sequencing. Analysis RFLP of COI-II and sequences of 750 bp of 3' end of COI gene showed that there are two main groups within specimens. Rate of sequence variations were up to 5 percent which is far exceeding than inter-population or -species variation. It is suggested that presumably this species is a group species. The results of this study which is the first molecular study on *An. superpictus*, needs to be confirmed by further molecular, cytological, and morphological studies and checked with available epidemiological data to reveal the relationship between these haplotypes and malaria transmission in each locality.

Micromalthus debilis LeConte and its varied symbionts

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The wood-boring telephone pole beetle, *Micromalthus debilis,* is the only member of the primitive family Micromalthidae. It has fascinated entomologists for years because of its unique life history features such as haplodiploidy, parthenogenesis, paedogenesis, and matriphagy. Many aspects of its biology still remain a mystery due to complications in rearing and manipulating the fragile individuals and broods. In spite of the scarce experimental data available, speculations on the evolutionary biology of this insect have been plentiful; for example, obligate matriphagy by males has been linked to the presence of a non-functional nutritional endosymbiont suggesting that *Micromalthus* is a case of Hamilton's hypothesis about the evolution of haplodiploidy, and the high cost of producing males has created a selective advantage for cyclic thelytoky. Using microscopical and molecular tools we are screening the symbiotic load of the different larvae and adults of *Micromalthus.* Several symbionts have been identified so far, including renowned reproductive parasites. We discuss the life history traits and evolutionary biology of this beetle in relation with our results and the comprehensive early literature.

Rapid cold hardening increases aphid survival at low temperatures.

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Most studies on insect cold hardiness have focused on seasonal adaptations occurring within the overwintering stage of an insect's life cycle, triggered by decreasing temperature and photoperiod from summer through autumn to winter. Due to their short generation times, an aphid that enters winter is unlikely to be the same individual that survives until spring. For this reason, although aphids are able to cold harden when reared at lower temperatures (10° and 5°C), the ability to rapidly cold harden may be relatively more important than in other longer-lived overwintering insects, where seasonally-induced cold hardening is the dominant response. This study assesses the effects of sub-zero temperatures on the mortality of nymphs and adult *S. avenae*, provides evidence of rapid cold hardening in *S. avenae* and investigates the ecological costs of rapid cold hardening in terms of effects on development, longevity and fecundity. The hypothesis that rapid cold hardening can be induced during the cooling phase of natural diurnal temperature cycles is supported. Rapid cold hardening increases survival and extends the lower lethal temperature, allowing aphids to acquire maximum cold hardiness when it is most needed.

Using geometric morphometrics to investigate wing dimorphism in males and females of hymenoptera – a case study based on the genus *Tachysphex* (Sphecidae: Larrinae)

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Traditional morphometrics are currently complimented by an additional statistical method, namely geometric morphometrics which is based on the quantification of shape using *x,y* coordinates. It is characterized by the obtaining of an exact geometric description of the morphology, and the ease to visually determine which landmarks are responsible for differences/similarities. The amount of variation between the forewings of males and females of the genus *Tachyshpex* (Hymenoptera) is investigated here. The research investigates the extent of morphological similarities/differences between males and females of the same species, in order to make recommendations regarding inclusion of both sexes in a systematic or geometric morphometrics study. Mantel statistics revealed a good correlation between the male and female dataset, and although visual analysis of the RWA plots of the two sexes indicated significant correlations between sexes of the same species, differences in the position of the species clusters suggests that sexual dimorphism in wings are present. Results from the thin-plate spline analysis of the mean shape of the male and female dataset revealed which landmarks are responsible for the differences. It is concluded that, in an analysis of members of a genus, only one of the sexes be used in the analysis, as small-scale differences may influence the results.

Glue protein in *Drosophila*: protein with an obscure function?

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Perusal of literature from the first description of larval salivary gland secretory protein (Fraenkel & Brookes, 1953) in *Drosophila* namely "Glue Protein", to the recently published article suggests that the glue 'helps' pupa to 'fix' to a substratum. Several lines of genetic and biochemical investigations on this tissue specific and stage specific protein in different species/subspecies of *Drosophila nasuta* subgroup has revealed that (a) abundant glycosylated protein is synthesized (b) most protein fractions are synthesized by co-dominant X-chromosomal gene/s (c) SDS-PAGE mobility differences of major glue fractions probably reflect differences in amino acid composition (d) there is exuberant SDS-PAGE pattern polymorphism (e) there is heteroselection of X-chromosomal glue gene/s in females in the natural populations of *D. nasuta nasuta* and *D. sulfurigaster neonasuta*. Morphometric, quantitative and behavioural studies involving several *Drosophila* species revealed absence of correlation between larval/pupal/gland size and the quantity of secretions. Species producing abundant glue prefer to pupate in the medium, those producing lesser quantities pupate on solid surface, while those producing negligible quantities, pupate in cotton plugs. These results suggest that the larval salivary gland secretions in *Drosophila* have other functions, in addition/rather than to help the pupa to 'fix' to a solid surface.

The evolution of sexually-selected traits in the sand cricket, *Gryllus firmus*

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Sexually-selected traits typically consist of components of morphology, physiology and behavior. This is particularly evident in Orthoptera where males call to attract females. Male call song is determined by the morphology of the sound-generating structure, physiological costs of calling and female behavior. In this talk I explore the evolution of these components in the cricket, *Gryllus firmus.* I discuss the genetic basis of the morphological components that generate the song, the song components themselves and the underlying physiology. I show that female choice is determined by both presence/absence of song and by song components. Because of the physiological costs of calling, macropterous males call substantially less than macropterous males and attract fewer females. Thus sexual selection can act not only on the components of the traits involved in female attraction but also on traits necessary for dispersal and hence can potentially have far-reaching affects on fitness. A common garden experiment using three geographically separate populations shows that the components underlying female choice vary among populations. Further, comparison of female choice in a population maintained in the lab for twenty years shows that this choice is stable over time, suggesting that it is maintained due to pleiotropy and not linkage disequilibrium.

The evolution of genetic architecture in response to environmental change

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Environments are highly variable in both space and time. Insects, being ectothermic are particularly sensitive to changes in their thermal environment. Evolutionary response to changes in the thermal environment are contingent on the presence of available additive genetic variation and the direct effect of the environment on the expression of this variation. I discuss two methods of investigating these factors: the experimental manipulation of temperature and the analysis, using a common garden experiment, of genetic variation along clinal gradients. For the experimental work I describe work with the cricket *Gryllus firmus*, showing that changes in temperature can have highly significant effects on phenotypic expression but relatively little on the genetic variance-covariance matrix. However, there are differences between wing morphs suggesting that these morphs show differential canalization responses to temperature change. For the work on geographic variation I describe common garden experiments conducted on two species of grasshopper (*Melanoplus* spp). These experiments show that the clinal variation in phenotypic traits is matched by clinal variation in genetic variation and that the differences among populations is concordant with previous phylogenetic analysis.

Reinvestigation of the mechanism of the female–biased sex ratio in *Wolbachia*–infected *Ostrinia furnacalis* (Lepidoptera: Crambidae)

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Wolbachia-infected females of *Ostrinia furnacalis* and *O. scapulalis* produce all-female offspring. The all-female production was interpreted as feminization of genetic males, since the antibiotic elimination of *Wolbachia* resulted in all-male offspring. However, in *O. scapulalis*, observation of the sex chromatin suggested that male-killing is the cause of the all-female production and that female-specific death is the cause of the all-male production after *Wolbachia* elimination. In the present study, we observed chromosomes of infected and cured insects of *O. furnacalis* to reinvestigate mechanism of the all-female production in this species. Infected female adults were found to be genetic females based on the presence of the female-specific sex chromatin. Both genetic males and females were included in the hatched larvae in the broods derived from infected females. Likewise, both genetic males and females were found in the hatched larvae in most broods derived from cured females. In contrast, the last instar larvae derived from infected females were in the all-female condition, and those derived from cured females were in the all-male condition. These results suggested the occurrence of opposite sex specific lethality in *Wolbachia*-infected and cured females of *O. furnacalis*, as suggested in *O. scapulalis*.

Paropsis atomaria (Olivier) (Coleoptera: Chrysomelidae):The significance of genetic and ecological diversity in a wide-ranging insect species

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The *Paropsis atomaria* complex currently consists of three described species, *P. atomaria*, *P. deboeri* and *P. elytrura*. While the latter two species have restricted distributions (to Tasmania and South-west Western Australia, respectively), *P .atomaria* is widely distributed in temperate, sub-tropical and tropical eastern Australia. The species are all *Eucalyptus* herbivores and are morphologically and behaviourally similar. This paper reports on a detailed investigation of both inter- and intra-specific relationships within the complex. Using molecular, morphological and ecological approaches the specific status of the three named taxa, as well as population level differentiation within the widely distributed *P. atomaria,* is being investigated. Special attention is paid to *P. atomaria* as it is an important forestry pest and there is potential that elucidation of population characteristics may reveal genetically distinct populations, possibly cryptic species, within the taxon. Assessment of the mitochondrial genome (COI, 550 b.p.) revealed approximately 9% differentiation between mainland *P. atomaria* and *P. deboeri*, whilst demonstrating a maximum of approximately 1.5% differentiation within *P. atomaria.* This supports current morphological classification. Such a multi-disciplinary, holistic approach will ultimately provide a robust foundation for the understanding of species boundaries and population dynamics in this economically important complex.

The water insoluble core of the silk filament is a polymer of proteins produced in the posterior section of silk glands. Polymerization into a fiber occurs via weak molecular interactions between amino acid residues that are arranged in specific repeats of a large protein called heavy chain fibroin (H-fibroin). Short non-repetitive ends of the H-fibroin molecule are similar in different Lepidoptera but the repetitive region, which accounts for about 95% of the H-fibroin length, has diversified under selection pressure for diverse physical properties of the silk and efficient deployment of available nutrients in its production. In some cases, silk may be used as a temporal depot of energetically rich and essential amino acids. In the "bombycoid" type of H-fibroin, the repeats vary in length and include concatenations of short crystalline motifs such as GlyAlaGlyAlaGlySer in the silkworms (Bombycidae) and stretches of about ten alanine residues in the silkmoths (Saturniidae). Owing to iterations of simple motifs, some amino acid residues of adjacent peptide chains are always in positions allowing intermolecular interactions even when the repeats do not exactly match. H-fibroins of the "pyraloid" type lack the strings of simple motifs and consist of complex repeats encompassing in some cases more than one huned amino acid residues. The uniformity of repeats throughout the H-fibroin molecule facilitates precise alignment of the widely distributed amino acid residues that form intermolecular links.

In most Lepidoptera, two small proteins called light-chain fibroin and P25 interact with H-fibroin in fibre formation.. The small components have been lost in the silkmoths (Saturniidae), whose silk fiber core consists of H-fibroin alone. Analysis of the dominant gene transcripts in the silk glands of two species representing two out of three caddisfly suborders, revealed presence of a product that is Gly-rich and may be regarded as a homologue of H-fibroin with very specific repeat structure. Another abundant mRNA seems to represent a homologue of light-chain fibroin. No sequence resembling the P25 component of lepidopteran silk was detected.

The core of silk fibre is enveloped by several sericins that are derived from two or more genes expressed in the middle silk gland section. Differential splicing and probably also different extent of glycosylation provide for the diversity of sericins that serve as a glue for fiber adherence. There are at least six different sericins in the silks of the examined Lepidoptera and apparently a similar number in the Trichoptera. Sericins are rich in serine and some of them consists largely of species-specific repetitive motifs.

Additional silk proteins of the lepidopteran silk include structurally diversified seroins and two types of relatively conserved proteinase inhibitors. These proteins are largely produced in the middle silk gland section prior to cocoon spinning and are believed to be responsible for the cocoon resistance to predators, molds, and microbes. Homologous proteins have not so far been detected in the caddisflies. Caddisfly silk contains several components whose relationship to the silk proteins of Lepidoptera remains to be elucidated. These caddisfly-specific silk proteins, which range in size from 13 to nearly 40 kDa, contain repetitive regions and structurally fall into three categories. One is characterized by high contents of Gly, Pro, Gln, and Trp, the second contains four short repeats rich in Cys, and the third includes repeats rich in Pro, Thr, and Lys. Some of other proteins encoded by the identified mRNAs could also be secreted into silk. They include a serpin (401 amino acids), a protein (140 residues) characterized by regular spacing of Glu, a protein (104 residues) with seven Cys residues, and a ProAla rich protein (61 residues). Taken as a whole, available data demonstrate that only the very basic composition of the caddisfly silk is similar to that known from Lepidoptera. Structures of the major proteins are different and several auxiliary compounds seem to be specific for Trichoptera. (The study was supported by grant A5007402 from the Grant Agency of the Czech Academy of Sciences).

Adaptation to environmental gradients.

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Latitudinal clines are widespread and provide a natural framework within which to examine natural selection. Repeated clines in different geographic areas or in related taxa strongly implicate the occurrence of natural selection. Clinal variation at the phenotypic level therefore provides an opportunity to examine the way in which genetic variation may also be influenced by natural selection. Despite the widespread occurrence of latitudinal clines, there are few empirical data that address the question of how quantitative genetic variation varies along such clines. We have combined comparisons of clinal populations of *Drosophila serrata* collected from the eastern coast of Australia with laboratory natural selection experiments to examine the genetic basis of adaptation to environmental gradients. We show that clinal selection under natural conditions does result in changes in both the additive and nonadditive genetic variances. Furthermore, we show that adaptation to clinal environments may involve a number of distinct genetic responses along the length of the cline, the complexity of which may not be fully revealed by focussing primarily on populations at the ends of the cline.

Metabolism of the sub-Antarctic caterpillar *Pringleophaga marioni* during cooling, freezing and thawing

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Although general models of the processes involved in insect survival of freezing exist, there have been few studies directly investigating physiological processes during cooling, freezing and thawing, without which those models remain hypothetical. Here we use open-flow respirometry to investigate metabolism of the freeze-tolerant sub-Antarctic caterpillar *Pringleophaga marioni* Viette (Lepidoptera: Tineidae) during cooling, freezing and thawing, and to compare animals exposed to non-lethal (-5.8 °C), and lethal (-6.0 °C, after which caterpillars are moribund for several days; and -18 °C, after which caterpillars are completely unresponsive) freezing stress. We found a large decrease in metabolic rate (that is not associated with freezing) at -0.6 ± 0.1 °C, and calculated a Q_{10} of 2.14×10^3 at this breakpoint. This breakpoint is coincident with the critical thermal minimum, and is hypothesised to be a metabolic manifestation of the latter. Caterpillars froze at -4.6 ± 0.1 °C, and had non-zero metabolic rates when frozen. Post-thaw, metabolic rates were lower than pre-freezing measurements. Post-thaw metabolic rates did not differ between temperatures that did and did not kill the caterpillars, which suggests that mortality may be a result of a breakdown in processes at the organismal, rather than cellular, level of organization.

Evolutionary implications of infection with parthenogenesis inducing *Wolbachia*

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The presence of Parthenogenesis Inducing (PI) *Wolbachia* infected individuals can have a strong impact on both the infected individuals as well as on the uninfected individuals present in a population. Two situations exist, the infection remains at some equilibrium or the infection goes to fixation. In this presentation I will present a model showing that an equilibrium infection frequency can only exist at a low level of infection if there are some factors present in the population that counteract the *Wolbachia* by modifying its expression /transmission (suppressor), or when other genetic elements are present that exploit the female biased sex ratio caused by the infection. If populations have gone to fixation for the infection with PI *Wolbachia* this generally is accompanied by a change in the females mating behavior and/or fertilization behavior. These changes are such that females from infected lines are unable to successfully mate and/or fertilize their eggs, whereas males derived from these same infected lines (through antibiotic treatment) are still functional and capable of fathering offspring.

Does environmental stress increase genetic structure in the springtail *Orchesella cincta***?**

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Environmental stress can alter the genetic composition of natural populations by directional selection towards tolerant genotypes, by causing bottlenecks and by changing gene flow to and from stressed locations. Especially when gene flow is low, environmental stress is expected to increase levels of genetic differentiation among populations. We examined the effect of environmental pollution on population genetic structure for the soil dwelling species *Orchesella cincta* **by comparing Isolation by Distance relationships. First, population genetic structure of sixteen NW European populations was estimated using six polymorphic microsatellite loci. A significant Isolation By Distance relationship was revealed, indicating gene flow is relatively high in this small soil arthropod species. Secondly, comparisons between stressed and pristine populations versus pristine populations revealed no differences in Isolation by Distance relationships. Our results indicate environmental pollution does not play a significant role in shaping population genetic structures within this species, most likely due to its migration capacity. Gene flow might, by filling up reduced genetic variability in stressed populations, be an evolutionary force maintaining versatility of stressed populations.**

"Effects of eugregarine parasites on adult longevity in the polymorphic damselfly *Mnais costalis* Selys"

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Males of *M. costalis* show a genetic polymorphism linked to mate-securing strategies, typically existing as orange-winged territorial fighters and clear-winged nonterritorial sneaks (the former are larger in size) (Tsubaki 2003). Orange-winged males have a higher daily mating rate, however, their reproductive life span and longevity is less than that of clear winged males. As a result, the estimated lifetime reproductive success is the same (Tsubaki *et al.*, 1997). In this paper I will compare the survival cost of eugregarine infection for the two morphs looking at the relationship between the abundance of mid-gut parasites and the survival of field-collected males in captivity. Orange-winged males suffered higher parasite infection and showed shorter longevity than clear-winged males when they were fed at lower levels than the natural feeding rate, or when they were deprived of food. In contrast, morph longevity did not differ when they were fed until satiation. The survival of damselflies was negatively correlated with the parasite abundance except when damselflies were fed until satiation. The results suggest that the abundance of eugregarine parasites exerts a substantial cost, which is associated with the maintenance of alternative mating strategies, together with the higher developmental cost and energy expenditure of the fighter morph.

An evolutionary view of the sex communication in *Melittobia* spp.

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Melittobia are ectoparasitoids of late larval and pupal stages of several species of solitary bees and wasps. Females locate their hosts on which they will lay their eggs, and will represent the only nutritional resource to the developing larvae. Sex dimorphism is remarkable and males will fight to death to eliminate competing males as they mate emerging females. Males remain in the confines of the host's nest they developed in, while females may disperse to locate new hosts. Mating occurs after an elaborate courtship behavior, in which a male produced sex pheromone is involved, and is supposed to play an important role in mate recognition. We will describe the differences in the male pheromone of 4 species of *Melittobia* and evaluate the attractiveness of females to interspecific males, and discuss the role of the male sex pheromone and the cuticular hydrocarbon composition of these 4 species in sex mate recognition. We will discuss the evolutionary process involved in the chemical communication of this parasitoid and provide supportive molecular data.

The sociobiology of plant defense

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"Social context" can inflate or diminish the evolutionary success of a resistance gene by making genetic variance in fitness density/frequency-dependent. Three models illustrate. First, a plant growth model shows that intraspecific competition increases the benefit of resistance when attack levels are high (resistant plants usurp resources that damaged susceptible plants need for recovery), but also increases the costs when attack is absent (undamaged susceptible plants outgrow resistant ones). Next, a game theory model shows that the fitness benefit of resistance can vary through the interaction of enemy behavior with local genotype frequency: susceptible genotypes can gain associational resistance when at low frequency if enemies can move freely to more palatable patches; however, if enemies are restricted to single patches, the susceptibles are eventually found. Finally, plants with high allocation to resistance flower late, causing assortative mating for resistance under low herbivory. Heavy attack retards susceptibles, bringing them into flowering synchrony with resistants. A population genetic model shows that assortative mating inflates genetic variance during zero-herbivory periods when costs exceed benefits, but not during high herbivory periods when benefits exceed costs. This causes asymmetric selection responses that prevent fixation of resistance even if average benefit exceeds average cost.

The role of the hedgehog pathway in polyembryonic development

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Copidosoma floridanum is a polyembryonic parasitoid wasp that produces over 1000 individuals from a single egg. This clonal proliferative process is remarkable for a number of reasons but primarily because this insect system has evolved clonal proliferation as an adaptive strategy; therefore, understanding how cellular proliferation can occur without causing disease in this organism will increase our understanding of how the cell cycle is regulated in animals and how it might be possible to mimic this natural system. The Hedgehog (HH) signal transduction cascade is a candidate gene pathway that may be regulating proliferation within the dividing cells. Among metazoan taxa, one function of HH is to regulate cellular proliferation during development. Within *Drosophila,* for instance, HH has been shown to control the proliferation and differentiation of somatic stem cells and progeny within the ovary. To determine whether HH is implicated in polyembryonic proliferation in *C. floridanum*, we cloned the C. floridanum HH gene. Ultimately, we are interested in determining the role the *C. floridanum* HH signal transduction pathway has in polyembryonic proliferation. We predict that in areas of active cellular proliferation, high levels of HH will be translated, while in areas with no proliferation, HH expression will be absent.

The genetic basis of divergent gene expression in *Drosophila*

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Differences in gene expression play a central role in evolution. Both *cis*- and *trans*-regulatory changes contribute to divergent gene expression, but their respective contributions remain largely unknown. Using closely related *Drosophila* species, we examined *cis*- and *trans*-regulatory changes underlying interspecific expression differences. Functional *cis*-regulatory changes were detected by comparing the relative abundance of *D. melanogaster* and *D. simulans* transcripts in F_1 hybrids, whereas differences in *trans*-regulatory activity were inferred by comparing the ratio of allelic expression in hybrids to the ratio of gene expression between species. We found that interspecific expression differences were almost always accompanied by changes in *cis*-regulatory function. In addition, *trans*-regulatory divergence affected approximately half of the genes we examined. These data contrast with recent studies of intraspecific gene expression, which found that expression variation was caused primarily by differences in *trans*-regulation. Distinct patterns of *cis*- and *trans*-regulatory changes appear to underlie expression differences within and between species.

Phenotype matching in wild parsnips and parsnip webworms: costs and consequences.

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The coevolutionary consequences of present-day interactions between plants and their insect herbivores may vary among populations as envisioned in the Geographic Mosaic Model of Coevolution. The interaction between wild parsnip, *Pastinaca sativa*, and its principal herbivore, the parsnip webworm (*Depressaria pastinacella*), is mediated by the amounts of furanocoumarins produced in the plant and the ability of the insect to detoxify them. Within 20 midwestern U.S. populations, trait frequencies in parsnips frequently were matched by trait frequencies in webworms. Exceptions were found in populations of parsnips with high xanthotoxin chemotypes, which are difficult for webworms to utilize, and in regions where an alternative host, *Heracleum lanatum*, which has a different furanocoumarin profile from wild parsnip, is abundant. The maintenance of more susceptible genotypes of wild parsnip in all populations is presumably due to the high cost of furanocoumarin production, while mismatching in alternate host populations is likely due to the cost of adaptation by webworms.

Use of thermal data as an establishment screen for non-native biocontrol agents in the UK

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Most predatory insects and mites used in glasshouse biocontrol in the UK originate from tropical and semi-tropical climates. For this reason, the licensing system for the introduction of non-native species has operated under the assumption that winter would act as a natural barrier to the establishment of such species outside of glasshouse environments. This view has been challenged by the establishment in the wild of the predatory mite *Neoseiulus californicus* and the discovery of the predatory mirid *Macrolophus caliginosus* outside of glasshouses in winter. Whilst the impact of these species on native ecosystems is unknown, their establishment is considered undesirable. This paper describes a series of experiments used to determine a range of thermal characteristics (developmental threshold, day-degree requirement per generation, supercooling point, lethal times and temperatures, field survival) of four non-native biocontrol agents. The comparative data provide a retrospective ecophysiological explanation for the establishment of *N. californicus* and *M. caliginosus,* and also indicate that *Delphastus catalinae* and *Eretmocerus eremicus* would not survive outdoors in the UK under current climatic conditions. The experimental protocol applied to these species could be used as part of a routine screen for 'establishment potential' in the licensing system of non-native biocontrol agents in the UK.

Learning affects adaptation to novel environments: evidence from experimental evolution using *Drosophila*

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Verbal arguments and models suggest that the ability to learn should affect the evolution of genetically determined components of behavior. Depending on the author and model, learning should either hinder or facilitate the response to natural selection. These contrasting predictions had not been addressed experimentally. To fill this gap we exposed replicated populations of *D. melanogaster* to directional selection on preference for a novel oviposition substrate (orange or pineapple), with or without an opportunity to use aversion learning to decide which medium to avoid. This additional opportunity to learn affected the genetic response to selection, but the sign of this effect depended on the direction of selection: it slowed down the response to selection to prefer orange, but accelerated the response to selection to prefer pineapple. Furthermore, for both directions of selection, populations given an opportunity to learn evolved a higher learning ability. This last results indicates that learning may also be favored under directional selection, in contrast to the usual argument that learning is only adaptive if fitness consequences of a given behavioral action vary in time or space. The results can be explained as consequence of the way learning affects the genotype-phenotype map.

Molecular phylogeny of Antarctic Chironomidae investigated by 28S rDNA sequencing

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Two indigenous chironomid midges exist on the Antarctic continent, *Belgica antarctica* and *Parochlus steinenii steinenii*. A third, *Eretmoptera murphyi*, endemic to sub-Antarctic South Georgia, was recently accidentally introduced to maritime Antarctic Signy Island (South Orkney Islands). *B. antarctica* and *E. murphyi* share several morphological and ecological characteristics and are classified in the same subfamily, Orthocladiinae. This study aimed to clarify the relationships of *B. antarctica* and *E. murphyi* and to evaluate whether vicariance or dispersal have been the main factors shaping their current distributions. Nucleotide sequences from the large subunit (28S) ribosomal RNA were used to compare the two species with other chironomids (Orthocladiinae, Chironominae, Telmatogetoninae) collected in Tierra del Fuego. Drawing on regional geological reconstruction, we attempt to match the timing of cladogenetic and geological events. Our results suggest a monophyletic familial origin. However at subfamily level, relationships are unclear. This is not surprising, as Orthocladiinae represents the residue of Chironomidae after the removal of other better-defined taxonomic groups. To further clarify relationships within Orthocladiinae, comparisons are currently being made with a species of the genus *Smittia* that may be the closest ancestor to both *B. antarctica* and *E. murphyi*.

High local endemism of a saproxylic 'giant' springtail (Collembola) from south eastern Australia

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Comparative phylogeography can reveal processes and historical events that shape the biodiversity of species and communities. As part of a comparative research program, the phylogeography of a new endemic Australian genus and species of log-dependent (saproxylic) Collembola was investigated using a variety of genetic markers. Owing to the species' ecological specialisation and sedentary lifestyle, and the palaeoclimatic history of the study site (Tallaganda State Forest, NSW), we predicted genetic structure would correspond with five *a priori* microbiogeographic regions. Significant genetic differences exist among regional samples ('populations'). Closely related mitochondrial DNA haplotypes are co-distributed within a single region or occur in adjacent regions, nuclear allele frequencies are more similar among more proximate populations, and inter-population immigration is rare. The signature of historical vicariance was also evident: the spatial distribution of populations corresponds with four of the five putative microbiogeographic regions, and genetic contact zones are abrupt. The present-day distribution of genetic diversity in this species seems to have been impacted by three major climatic events: Pliocene cooling and drying (2.5-6 mybp), early Pleistocene wet-dry oscillations (1.2 mybp), and the more recent glacial-interglacial cycles that have characterised the latter part of the Quaternary (<0.4 mybp).

Morphometric, genetic & ecological variation in *Aphis gossypii*

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Aphis gossypii Glover was collected from a range of crop and non-crop plant hosts at various times during the year on Guam. *A. gossypii* was also collected from the Commonwealth of the Northern Marianas Islands in Saipan, Tinian and Rota, from the Republic of Palau islands of Koror and Babeldaup, from the Republic of the Marshall Islands, from five of the Hawaiian Islands, and from Japan, Canada, USA, and Syria. Aphid/plant host associations were determined. Morphometric analysis of slide-mounted specimens using principal components analysis suggested that aphid variation was most closely related to geographic provenance, followed by host plant. However, results from microsatellite flanking region sequence analysis (MFRS) performed on aphids from the same collections indicated that genetic variation was most closely related to the host plant from which the aphids had been collected. Field observations made on Guam suggest that the physical appearance of aphids may be altered by altering the host plant upon which the aphids develop. Collecting and analysis of variation in *A. gossypii* populations from various sites continues.

Comparison of *mariner*-like elements among *Bombyx mandarina* individuals inhabiting east Asia in the light of the segregation of *B.mori* and *B. mandarina*

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"Individuals of *Bombyx mandarina* were collected from three area of east Asia, Japan, Korea and China, observed for chromosome number (*n*) and subjected to PCR against their genomic DNA with the inverted terminal repeats of *Hyalophora cecropia* MLE as a primer. Each specimen gave several bands with different sizes. The 1.3-kb band, expected to cover a full-length MLE, was ubiquitous in all of the *B. mandarina* specimens as in *B. mori*. However, in some cases, the bands of *B. mandarina* were inserted with other sequences. The MLE making a complex was found to be situated in the same phylogenetic group as that of the previously analyzed MLE clone (*BmTNML*) from *B. mori*, indicating that the insertion had occurred in the ancestor of *B. mori* before the genomic segregation of *B. mandarina* between the Japanese/Korean types (*n* = 27) and the Chinese/Korean types (*n* = 28). One individual from China had a discrete 4.2-kb band containing a *BMC1* retrotransposon, which is a unique fragment detected in almost all of the silkworm races and strains. Thus, the 4.2-kb unit might be made spontaneously by crossing with *B. mori* after the segregation of *B. mori* genome.
"

Possible horizontal transfer of *mariner*-like sequences into some invertebrates including lepidopteran insects, grasshopper & a coral among Japan islands

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Mariner-like elements (MLEs) were amplified by PCR against genomic DNAs from several Japanese insects including lepidopteran lepidopteran species, using the inverted terminal repeats (IR) of the *Hyalophora cecropia* MLE as a primer. Clones thus obtained were of a size of about 1.3 kb, and expected to contain the full-length MLE. These clones were highly similar to each other and classified into the *Cecropia* subfamily of MLEs. The coral MLE was found to have a complete ORF coding for transposase, a situation similar to that previously found in the Emperor moth, *Attacus atlas*, from the Ryukyu Island. These findings, together with the fact that all of the insects and coral species that exhibited a positive signal for the full-length MLE were collected in relatively close regions around Japan, indicated that the horizontal transfer of MLEs has taken place even among phylogenetically remote organisms. We will also report the results regarding as to the transposition activity from one of these type of *mariner*-element isolated form *A. atlas* using the *Bombyx* cultured cell.

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Development of males in thrips – a bacterial decision

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In thrips the mechanism of reproduction usually involves either arrhenotoky where females develop from fertilized and males from unfertilized eggs, or thelytoky where females hatch from unfertilized, diploid eggs. Thysanoptera are the only insect order besides Hymenoptera showing a haplodiploid reproduction mode. To find out more about the mechanism determining sex in thrips, several thrips species were tested for the presence of parthenogenesis-inducing bacteria of the genus *Wolbachia* using specific 16S rDNA primers. Tested species show either arrhenotoky (*Frankliniella occidentalis*, *Echinothrips americanus*) or thelytoky (*Parthenothrips acaenae, Hercinothrips femoralis*), but *Thrips tabaci* is arrhenotokous in some areas and thelytokous in others. The thelytokous species *H. femoralis* and *P. acaenae* possess *Wolbachia*, and presence of this bacterium in *H. femoralis* was shown to be responsible for thelytoky since male offspring could be generated by treating females with antibiotics. After mating, sperm was found in the spermatheca of female *H. femoralis*, and mated females produced only female progeny. The arrhenotokous species *E. americanus* also harbours *Wolbachia*, but the bacterium was not detected in either *F. occidentalis* or *T. tabaci*.

Reproductive isolation & genitalic divergence in *Ciulfina*: a praying mantid genus from tropical Australia

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Populations of animals can be reproductively isolated from one another at a variety of levels: from precopulatory species recognition through to hybrid dysfunction. Genitalic characters are often used to define divergent populations as distinct species, particularly in the insects. However, the correlation between genitalic divergence and reproductive isolation in insect species is rarely tested. Mantids in the genus *Ciulfina* are small trunk-dwelling predators found in high densities in a variety of forest types throughout coastal Queensland, Australia. The degree of polymorphism found in both the male genitalia, and sequences of the ITS region of ribosomal DNA in *Ciulfina*, suggested that particular populations were reproductively isolated from one another. Using choice experiments, we found that males were unable to differentiate between females from their own population and those from populations that were genetically different. Once copulation was attempted, however, the duration and mechanics of interpopulation matings varied from those of intrapopulation matings. Most importantly, populations with some level of sympatry/parapatry were more effectively isolated from one another than entirely allopatric populations, suggesting that genitalia do serve an important role in species reinforcement. These results have implications for our understanding of the evolution of animal genitalia, and of the role of genitalic divergence in reproductive isolation.

Comparative evolutionary ecology of limestone & lava caves in tropical Queensland

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Recent discoveries of obligate cavernicoles from tropical caves and lava tubes have opened new areas for research. The Chillagoe limestone caves and Undara lava tubes, 100 kilometers south, provide exceptional systems to compare evolutionary ecology between limestone and lava caves since their communities evolved from the same source fauna within the same climatic regime. The 5 to 25 million year-old limestone caves formed slowly by solution. The Undara lava tubes formed within a 190,000 year-old eruption, but intermittent volcanism has maintained interconnected cave habitats for possibly five million years. Despite the very different substrates and modes of formation, the food resources and physical environments within the caves are similar. Both cave types contain numerous passages with different physical shapes and environments, allowing multiple comparisons in environmental ecology. The adaptations and distribution of cave species are correlated with the physical environment rather than with cave age or type, with the most cave-adapted species being restricted to the harshest environment. Over 40 obligate cavernicoles are known, but the fauna is potentially much larger, especially given the vast number of uninvestigated caves. Obligate cavernicoles evolve to exploit environments stressful for surface animals wherever suitable habitats and resources have been available for evolutionary time.

Identification of source populations of invasive insects: implications for biological control, monitoring, and quarantine

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Invasive species cause severe economic and ecological damage in many regions around the world. Knowledge of an invasive species' origin benefits inspection and quarantine efforts and focuses the search for biological control agents. Cooley spruce gall adelgid (CSGA), *Adelges cooleyi*, is a native pest in the US that provides an excellent model organism for studying population genetics of invasive exotic species. The native range of CSGA resembles an inverted 'V', extending northward along the Rocky Mountains into Canada and back down the West Coast into northern California. CSGA has invaded the eastern US through an unknown number of introductions during the 20th century. Here, we report efforts to identify sources of CSGA populations introduced into the eastern US. Comparison of mitochondrial and nuclear DNA sequences among source and introduced populations revealed that all east coast introductions originated from one region within the insect's extensive native range. As expected by theory, genetic variation was decreased in introduced populations relative to populations from the insect's native range. This work demonstrates a successful application of molecular genetic tools to determine the origin of invasive species and aspects of population genetic structure, thus facilitating the development of sound, sustainable pest management strategies.

Inbreeding leads to changes in allometry between egg size and larval characters in an eriosomatine aphid (Aphididae)

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A number of studies indicate that inbreeding has deleterious effects on the viability and reproduction of organisms, but it is still unknown what mechanisms are involved in the expression of deleterious effects. Because the effects of inbreeding are expressed from the early stage of development, it is necessary to focus on hatched larvae to understand the phenotypic effects of inbreeding. Aphids are useful for studying inbreeding effects because in aphids (1) it is easy to cross males and females of the same clone (that is, selfing) and (2) hatched larvae can be used for detailed morphometric analyses. Using the aphid *Prociphilus oriens*, this study prepared eggs resulting from outbreeding and selfing and compared the allometry between egg size and morphological characters of hatched larvae between outbred and selfed groups. The allometry of larval characters against egg size varied greatly between outbred and selfed groups. Selfed larvae had significantly larger allometric coefficients for appendages than did outbred larvae, showing that inbreeding accelerated the growth of appendages. However, the growth of gonads was suppressed in selfed larvae. It is suggested that inbreeding relaxed developmental canalization on larval appendages and resulted in reduction in gonad size via a trade-off between appendages and gonads.

Rapid chill coma recovery selection in *Drosophila melanogaster*

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Resistance to low temperatures can vary markedly among invertebrate species and is directly related to their distribution. Despite the ecological importance of cold resistance this trait has rarely been studied genetically. One measure of cold resistance that varies geographically in *Drosophila melanogaster* and that is amenable to genetic analysis is chill-coma recovery. *D. melanogaster* have been selected for increased recovery time following exposure to 0°C. Selected lines responded rapidly to the selection and showed an increased resistance to a broad range of low temperatures. They also had a lower mortality following a more severe cold treatment, indicating that a general mechanism underlying cold resistance had been selected. The selection response was independent of plastic changes in cold resistance because the selected lines maintained their ability to acclimate. Life-history and stress resistance tradeoffs were also investigated showing only a tradeoff with ageing and one stress resistance trait. Selected lines were genotyped for clinally varying genes/markers and show associations with some candidate genes suggesting these genes may have a role in cold resistance.

Predicting shifts in timing of winter moth egg in response to climate change

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Climate change can lead to mistiming in phenology between different components of food chains. For instance, winter moth (*Operophtera brumata*) egg hatch has advanced more than oak (*Quercus robur*) bud burst over the past two decades. The question is whether natural selection will lead to a restoration of synchrony between winter moth and oak. Using a half-sib design, we demonstrate here that egg hatching is a heritable trait, making adaptation through selection possible. We further use a model to predict the changes in reaction norm of egg hatching at different temperatures. This approach enabled us to predict the response in the next year, under different environmental conditions, to the selection in a given year. Due to selection, the reaction norm will change in intercept but not in slope: in future eggs will need more warmth before they can hatch, regardless of what the temperature is in a given year. Because of this, timing differences between egg hatch and oak bud burst will decrease rapidly. Changes in reaction norm in response to selection thus offer a possible way of restoring synchrony between different components of a food chain, winter moth and oak.

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Synchronous coadaptation in an ancient case of herbivory

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Coevolution has long been considered a major force leading to the adaptive radiation and diversification of insects and plants. A fundamental aspect of coevolution is that adaptations and counteradaptations interlace in time. A discordant origin of traits long before or after the origin of the putative coevolutionary selective pressure must be attributed to other evolutionary processes. Despite the importance of this distinction to our understanding of coevolution, the macroevolutionary tempo of innovation in plant defenses and insect counterdefenses has not been documented. Molecular clocks for a lineage of chrysomelid beetles of the genus *Blepharida*, and their Burseraceae hosts were independently calibrated. Results show that these plant's defenses and the insect's counterdefensive feeding traits evolved roughly in synchrony, providing the first macroevolutionary confirmation of synchronous plant-herbivore coadaptation. The association between these two groups of organisms was determined to be about 112 million years old, the oldest age so far for a specialized plant-herbivore association.

Genetics and evolution of photoperiodism in temperate environments

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Photoperiodism constitutes the major physiological mechanism by which arthropods adjust the seasonal programming of development and dormancy at temperate latitudes. Dispersal of the pitcher-plant mosquito, *Wyeomyia smithii*, from southern latitudes to northern Canada following recession of the Laurentide Ice Sheet 20,000 - 8,000 years ago has involved the evolution of longer critical photoperiods, thereby tracking the shorter growing seasons at increasingly more northern latitudes. Despite directional and stabilizing selection during post-glacial range expansion, the heritability of critical photoperiod increases with latitude. We attribute this increase in heritability to the release of additive from epistatic variance during sequential founder events. During rapid climate change over the last 30 years, critical photoperiod has shifted towards shorter, more southern day lengths as growing seasons have become longer. This genetic shift in photoperiodic response is detectable over a time interval as short as five years. Faster evolutionary response has occurred in northern populations where selection is stronger and genetic variation is greater. Our results suggest that other species may be in the process of analogous evolutionary responses to altered seasonality and that the composition of future biotic communities may depend on the relative abilities of their constituent species to adapt to altered seasonal interactions.

Fluctuating asymmetry of wing pattern elements and wing shape, and its relationship to migration in the butterfly *Melitaea cinxia*

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When fitness depends on morphological traits, individuals that can develop the phenotype reliably should be more fit than those, which cannot. Fluctuating asymmetry (FA; minute differences between the left and right side of a bilateral trait due to developmental errors) has therefore been used to predict the fitness of individuals in natural and sexual selection studies. In this talk we discuss how the FA of wing pattern elements and shape of the wings of both the fruitfly *Drosophila melanogaster* and the butterfly *Melitaea cinxia* can be used to obtain information on how development is stabilized and how useful FA is in a natural selection experiment. In the butterfly species a mark-release-recapture experiment was carried out to obtain information on the relationship between FA of wing pattern elements and shape, migration, past and present population sizes, dispersal, survival and mating success. The results will be discussed taking recent results into account on the mechanisms of developmental stability as obtained with *D. melanogaster*.

The effect of a resistance gene on developmental stability and fitness in a flea beetle, *Phyllotreta nemorum*

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A resistance (R-) gene enables the flea beetle *Phyllotreta nemorum* L. (Coleoptera: Chrysomelidae: Alticinae) to use the otherwise toxic *Barbarea vulgaris* ssp. *arcuata* G-type plant as a host plant in Denmark. In this study, the pleiotropic effects of the dominant R-allele at a single autosomal gene, originating from the Danish region of Kværkeby, were investigated in an isogenic background from non-resistant beetles. The presence of the R-allele causes a high mortality at the early stages of development, when the toxic G-type plants cause the high mortality in non-resistant beetles. The presence of the R-allele has little or no effect, even under suboptimal conditions, on developmental stability (measured as fluctuating asymmetry of tibialength), body size, fecundity and longevity in the adult beetles. The distribution of the beetles in the field in Denmark will be discussed and its host plant use.

Contemporary evolutionary genetics: the breadth and depth of host race evolution in bugs on two continents.

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Several species of seed bug (the soapberry bugs: Rhopalidae) from the Americas and Australia are specialized on the plant family Sapindaceae ("the 'soapberry family"), including exotic introductions and escapes. Studies on species in both Hemispheres reveal morphological, physiological, behavioral and life history adaptations to host fruits and phenologies. Many of these adaptations display rapid or contemporary evolution (in 30-50 years or 100 generations) on introduced hosts; they include traits from mouthpart morphology to wing muscle enzyme physiology. We document this evolution in soapberry bugs from Australia (*Leptocoris*) and North America (*Jadera*), showing geographic variation within continents and among host plants. The genetics underlying contemporary evolution has been analysed in a North American species using hybrids between populations adapted to different hosts as well as artificial selection. The analyses demonstrate that additive and non-additive genetic variation in the various traits contributes to contemporary evolution. Further, they reveal expected and some unexpected genetic correlations and interesting evolutionary pathways to adaptation.

A macrophysiological perspective on cold and desiccation tolerance

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Macrophysiology, or the investigation of variation in physiological traits over large geographic and temporal scales and the ecological implications of this variation, is gaining increasing attention globally for several reasons. Here, I explore large-scale variation in cold tolerance and desiccation in insects and its implications. Perhaps unsurprisingly, rates of water loss vary in an environmentally consistent fashion, such that rates are lower in dry environments. This is true of both cuticular and respiratory water loss, even though the significance of the latter and its likely response to selection have been questioned. In several species it is clear that respiratory water loss is remarkably sensitive to environmental variation and adaptive variation thereof is not inconsistent with the data. Patterns in cold tolerance are more complicated, and reflect the variety of strategies animals can adopt to survive low temperatures. Nonetheless, two major patterns are emerging from the broad scale data. First, there is decoupling of upper and lower thermal limits. Second, there are pronounced and consistent differences in the way temperate species from the Northern and Southern Hemispheres respond to low temperatures. These major patterns not only provide insights into the mechanisms underlying insect responses to cold, but also have significant implications for understanding and predicting species responses to environmental change.

Costs of salicylate- and jasmonate-mediated induced responses in *Arabidopsis thaliana*

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Fitness costs of plant defenses to herbivores and pathogens are an important aspect of the evolutionary dynamics of plant resistance. Costs of defense have been increasingly reported in the literature, yet their mechanisms remain largely unexplored. Costs may arise from increased allocation of resources to defense production at the expense of growth, decreased assimilatory capacity in highly defended plants, or in the case of induced responses, negative interactions between defense pathways that compromise resistance to certain natural enemies. I explored costs of induced responses in several *Arabidopsis thaliana* genotypes that vary in their constitutive and inducible expression of resistance traits. In the absence of benefits, highly defended genotypes, such as the constitutive expressor of systemic acquired resistance mutant, *cep1*, had lower seed production than less defended genotypes, in general. Plants induced with either salicylic acid or jasmonic acid produced less seed than uninduced plants, and costs were quantitatively associated with the induction of such defenses as trypsin inhibitors and glucosinolates, coupled with decreased assimilatory capacity. In addition, jasmonate-mediated resistance to insects was compromised in plants simultaneously induced with salicylic acid due to inhibitory defense pathway crosstalk, and an inverse relationship between bacterial and insect resistance was demonstrated for several genotypes.

Forewing shape variation in the sand cricket *Gryllus firmus*

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Populations of the sand cricket *Gryllus firmus* are composed of two wing morphs. Long winged individuals (LW) have functional hind wings, are able to fly, and thus might disperse. Short winged individuals (SW) have vestigial hind wings and cannot fly. It is commonly considered that this dimorphism is maintained by a trade-off between flight capacity and fecundity. Here, we focused on the male forewing. Since males use their forewings to produce courtship songs, this structure affects fecundity. Additionally, it is likely that some genes jointly affect the two pairs of wings. Selection for flight efficiency on the hind wings should thus indirectly affect the forewings. This implies that the fore wings might be influenced by both trading off parameters. Using geometric morphometrics we analyzed the forewing shape variations both within and among morphs. Then, using a full sib design we estimated the genetic and non genetic components of shape variation. We discuss our results in terms of selection and developmental constrains.

Molecular responses to cold and desiccation

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Heat shock proteins (Hsps) are involved in responding to both low temperature and desiccation in flesh flies, but different sets of Hsps are involved in the responses to overwintering cold hardiness, rapid cold hardening, desiccation and rehydration. The use of RNAi to knock out Hsp70 has a dramatic effect on overwintering cold hardiness, but Hsp23 appears to be much more important for rapid cold hardening. Hsp23 and Hsp70 are both upregulated by desiccation, but two other Hsps, Hsp90 and Hsc70, are upregulated when a desiccated fly is rehydrated. The rehydration response is most similar to the Hsp response to cold recovery: Hsp90 and Hsc70 are upregulted in both cases. The deployment of different subsets of Hsps thus differs from the response to heat shock, where all are upregulated simultaneously. Microarray data indicates that a number of genes, in addition to the Hsps, are involved in the fly's response to low temperature.

Phylogenetic congruence of mealybugs and their P-endosymbonts

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Tight interactions between unrelated organisms such as is seen in plant-insect, host-parasite, or host-symbiont associations may lead to speciation of the smaller partners when their hosts speciate. Totally congruent phylogenies of interacting taxa have not been observed often but a number of studies have provided evidence that various hemipteran insect taxa and their primary bacterial endosymbionts have shared phylogenetic histories. Like other hemipteroids, mealybugs (Pseudococcidae) harbor multiple intracellular bacterial symbionts that are thought to be strictly vertically inherited. Thus co-divergence of hosts and symbionts is expected. Here, robust estimates of phylogeny were generated from four fragments of three nuclear genes for mealybugs of the subfamily Pseudococcinae, and a substantial fragment of the 16S-23S rDNA of their P-endosymbionts. Phylogenetic congruence was significant, with 75% of nodes on the two trees identical. Congruence was not perfect however, and other evolutionary outcomes including host shifts could not be ruled out. It is shown that incongruence may be influenced by uncertainty in phylogenetic estimation.

Effects of short-term spontaneous mutation accumulation for life history traits in grape phylloxera

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Mutation is the source of all genetic variation but the mutation rate, effects of new mutations, and their role in the maintenance of genetic variation are still subject to controversy. These parameters are important in models of the evolution of sex and recombination, the persistence of asexual populations, and the extinction of small populations. Most estimates have come from model organisms. Here, mutation accumulation experiments were conducted with 3 clones of grape phylloxera, *Daktulosphaira vitifoliae* Fitch, a gall forming herbivore of grapes, to estimate rate of input and effects of spontaneous mutation on life history traits. This is perhaps the first such experiment using a non-model organism of economic importance. Evidence was found that mutation rates are high enough to produce heritable genetic variation over short time scales, and that the effects of mutation on these quantitative traits are deleterious on fitness. Estimates of the parameters by the Bateman-Mukai method were within the range found in previous studies. Rapid accumulation of deleterious mutations suggest that for many pest species, adaptive response under agricultural conditions may depend more on standing variation from introductions than new mutation.

Insect antifreeze proteins

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Although they have been identified in various animals, plants, fungi and bacteria, the most active antifreeze proteins (AFPs) yet discovered are found in insects. In larvae of the freeze avoiding Pyrochroid beetle *Denoides canadensis* AFPs function to prevent freezing by (1) inhibiting inoculative freezing initiated by external ice across the cuticle, and (2) by inactivating internal ice nucleators, thereby inhibiting spontaneous nucleation and promoting supercooling. Two types of insect AFPs are currently known. Those of *D. canadensis* (DAFPs) consist of 12-and 13-mer repeating units in which certain positions (especially those with threonine, serine and cysteine residues) are highly conserved. Threonines, in particular, appear to be involved in the ability of the DAFPs to hydrogen bond to ice or ice nucleators. There are tissue and temporal variations in the expression of the family of 13 variants of the DAFPs, suggesting variation in function. Optimal function of the DAFPs requires the presence of low molecular mass enhancers (glycerol, etc.) and/or certain other protein enhancers. Also, certain DAFPs have a synergistic effect on other DAFPs. In Alaska, AFPs similar to DAFPs play a role in the ability of larvae of the Cucujid beetle *Cucujus clavipes* to supercool to extreme temperatures, in some cases below -80C.

Canalization and cryptic variation in *Drosophila melanogaster*.

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Canalization has been discussed a great deal with respect to evolutionary developmental biology, however there is a lack of consensus as to a precise definition of canalization itself. This has resulted in a large number of studies exploring different aspects of the same question with no common reference. I will discuss some methodological and statistical approaches to addressing questions of canalization, and cryptic genetic variation, and provide some empirical examples of how these can help to improve our understanding of canalization. Furthermore, I will summarize the results of a recent association mapping study designed to explore the genetic basis of cryptic variation.

Evolutionary and functional genomics of wing shape in *Drosophila melanogaster*.

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While most organisms vary considerably in aspects of shape, surprisingly little is known about the genetic basis of shape variation. Using the wing of *Drosophila* as a model system, I will present the results of a functional genomics study of wing shape, and how mutants in known wing patterning genes affect shape. Furthermore I will discuss the results of association mapping studies between natural polymorphisms in two genes, *Epidermal growth factor receptor* and *hedgehog*, with wing shape.

Diversity in the weapons of sexual selection: horn evolution in the beetle genus *Onthophagus*.

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Both ornaments and weapons of sexual selection frequently exhibit prolific interspecific diversity of form. Yet, most studies of this diversity have focused on ornaments involved with female mate choice, rather than on the weapons of male competition. With few exceptions, the mechanisms of divergence in weapon morphology remain largely unexplored. Here, I examine the evolutionary radiation of one type of weapon: beetle horns. I use partial sequences from one nuclear and three mitochondrial genes to develop a phylogenetic hypothesis for a worldwide sample of 39 species from the dung beetle genus *Onthophagus* (Coleoptera: Scarabaeidae), and then use these data to trace the evolutionary radiation of horns. Although this limited sample of one of the world's most species-rich genera almost certainly underestimates the number of evolutionary events, it reveals prolific evolutionary lability of these exaggerated sexually selected weapons (over 24 separate gains and losses of five different horn types).

Genotype to phenotype: physiological control of trait size and scaling in insects.

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For almost a century, biologists have used trait scaling relationships (bi-variate scatter-plots of trait size versus body size) to characterize phenotypic variation within populations, and to compare animal shape across populations or species. Scaling relationships are a popular metric because they have long been thought to reflect underlying patterns of trait growth and development. However, the physiological mechanisms generating animal scaling are not well understood, and it is not yet clear how scaling relationships evolve. Here we review recent advances in developmental biology, genetics, and physiology as they pertain to the control of growth of adult body parts in insects. We summarize four mechanisms known to influence either the rate or the duration of cell proliferation within developing structures, and suggest how mutations in these mechanisms could affect the relative sizes of adult body parts. By reviewing what is known about these four processes, and illustrating how they may contribute to patterns of trait scaling, we reveal genetic mechanisms likely to be involved in the evolution of insect form.

Impact of 1997-1998 ENSO-induced fires and logging on the genetic diversity of *Upadia theda* Felder (Lycaenidae) in Borneo.

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Drastic changes in population size as a result of disturbance can severely affect genetic variation. However, although severe disturbance is assumed to reduce levels of diversity, intermediate levels of disturbance, on the contrary, can lead to elevated levels of diversity. Here, we investigated the temporal variation in genetic diversity in populations of the butterfly species *Upadia theda* Felder (Lycaenidae), that were differentially affected by disturbance in East Kalimantan, Indonesia. We focused on the two main forms of disturbance that affect tropical rain forests, namely logging and ENSO-induced drought and burning. The populations were sampled over a four-year period in pristine, logged and burned areas before and after 1997-1998 ENSO-induced event. We assessed genetic variation within and between samples by analysing mitochondrial DNA sequences (mtDNA control region+ 5'end of 12S) and five microsatellite loci. Both molecular markers gave concordant results with respect to the temporal pattern of genetic diversity within disturbed areas: a significant loss of genetic diversity was observed in burned areas compared to pristine areas, but there was an increase of observed genetic diversity within logged areas. Gene flow between *Upadia theda* populations appeared, furthermore, to be highly restricted at the geographic scales assessed (less than 100 km). Importantly, our genetic diversity results are in line with previous published studies on temporal and spatial patterns of butterfly species richness.

Human-induced changes in organism distributions and the evolution of insect diets

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Human activities have caused many species to expand their ranges and humans have introduced organisms into many new and often very novel environments. These human-induced changes in organism distributions create natural experiments with which we can study ecological and evolutionary processes that otherwise could only be inferred indirectly. I examine the consequences recent changes in plant distributions for the evolution of insect diet. Using a model system of diet evolution is seed beetles (Coleoptera: Chrysomelidae: Bruchinae) I examine evolutionary constraints on adaptation, the role of life history plasticity, and the importance community composition in mediating diet expansion and adaptation.

Investigating the genetic basis of phenotypic variation in *Drosophila* egg shell patterning.

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Patterning of the insect egg shell provides an excellent system for exploring the molecular basis of phenotypic variation. In *Drosophila melanogaster*, two dorsal-anterior respiratory appendages are produced in response to signaling through the *Epidermal growth factor receptor* (*Egfr*). Previous empirical and theoretical work implicates *Egfr* function in both intra-specific variation for dorsal appendage spacing on the eggshell, as well as inter-specific differences in dorsal appendage number and location. To test the hypothesis that genetic variation in *Egfr* contributes to variation in egg shell patterning, we have made use of naturally occurring intra-specific variation for dorsal appendage spacing as a model quantitative trait. An initial association study suggests that two common polymorphisms in *Egfr* are involved in trait expression. We will report on our efforts to confirm the involvement of these SNPs as well as efforts to identify additional loci that contribute to dorsal appendage spacing.

Microbial diversity in termite guts and their role in mineral transformation of soil

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Termite a macro invertebrate act as soil engineers in tropical ecosystem modifying the environment in both biotic and abiotic components. Termites have a major effect on the physical properties, increases water holding capacity, control soil erosion and also improves the crop sustainability. Polysaccharide producing bacteria *viz., Azotobacter* and *Beijerinckia* isolated from the foregut of termites which are responsible to fix atmospheric nitrogen. Soil aggregation improves the physical parameters - porosity (40.3%), bulk density (1.33 g/cc), particle density and water holding capacity (39.8%), chemical parameters – increased organic carbon (0.098%) and available nitrogen (70 ppm). Incorporation of *Beijerinckia* sp. in acid soils provides additional attributes for crop production and environmental sustainability. The fungi *viz., Termitomyces* isolated from the fungal garden involved in the degradation of leaf litter in terrestrial ecosystem and also used as edible mushroom due to its medicinal value. Hence termites, are not only pests, but also play an important role in sustaining soil ecosystems.

Costs and benefits of glandular trichomes in *Datura wrightii*: why be Sticky?

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The relationship between costs and benefits of herbivore resistance predicts the evolution of resistance. *Datura wrightii* (Solanceae), is dimorphic for leaf trichome type. Some plants are covered with short, non-glandular trichomes ("velvety plants"), while others are covered by longer, glandular trichomes that secrete acyl sugars ("sticky plants"). Glandular trichomes confer resistance to many insects but not all. In the absence of herbivores, glandular trichomes are costly because sticky plants produce 45% fewer seeds than velvety plants. Sticky plants grow larger than velvety plants, and this may confer a competitive advantage. In a three-year field study, plant competition reduced plant seed production by 76 – 87% overall. Competition was asymmetrical and favored sticky plants because sticky plants produced 42% more seeds when competing with velvety plants than when competing with other sticky plants. Conversely, velvety plants produced 53% fewer seeds when competing with sticky plants than when competing with other velvety plants. Increased competitive ability is an additional potential benefit of the suite of traits associated with the production of glandular trichomes in *D. wrightii* and may contribute to the maintenance of the trichome dimorphism in natural *D. wrightii* populations.

The mating acceptance rate of female was higher for large males than for small males.
Males fought with other males over seeds using his fore legs, and the larger males won in all cases.
Females were attracted to some odour of male, and it was more effective when males were accompanied with seeds.

In this species, copulation interval is much longer in females, which makes OSR male-biased and competition over females severe. We conclude that males adopt Resource Defence Mating System, in which they defend seeds and/or area from other males, and attract females with some odour. Therefore, sexual selection favours larger male body size.

Wing shape, wing size and fitness in *Trichogramma* wasps

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Trichogramma wasps are tiny egg parasitoids widely released commercially to control moth pests. To evaluate the impact of wing shape and wing size on wasp fitness, we released wasps at a central point and compared the morphology of female wasps in a release sample with females that successfully located eggs of target hosts. Three species-host-plant combinations were evaluated: *T. carverae* attacking lightbrown apple moth (*Epiphyas postvittana*) on grapevines, *T.* nr. *brassicae* attacking *Helicoverpa* spp. on tomatoes, and *T. brassicae* attacking European corn borer (*Ostrinia nubilalis*) on corn. In each case, wing size was calculated as centroid size and wing shape was assessed using Procrustes superimposition. Wing size of wasps had an impact on field fitness in all cases, but in *T. brassicae* this association was complicated by the fact that wing size effects changed with distance from the release point and time since release. Wing shape had no impact on field fitness in *T. brassicae*, but did influence field fitness in *T. carverae* and *T.* nr. *brassicae*. Changes in wing shape associated with high fitness were different in the two situations. Rearing host influenced wing shape. In all cases shape effects appeared to be at least partly independent of allometry.

Clinal variation and the genetics of stress resistance

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Clinal variation for traits within species provide an outstanding resource to identify traits and genes under selection due to climatic factors. The most well known clines in insects are for body size, but clinal variation in a range of other traits has also been identified. One of the most intensively studied clines is the *Drosophila melanogaster* cline along the east coast of Australia where populations can be collected along a transect ranging from temperate Tasmania to tropical Far North Queensland. There are opposing patterns along this transect for resistance to cold and heat stress, as well as clinal variation for evasion of stressful winter conditions by altered patterns of egg production. We are now dissecting these patterns phenotypically and genetically to identify the physiological and genetic basis of adaptive shifts in these traits. QTL mapping following crosses between populations from cline ends indicate regions of the genome associated with traits, while QTL mapping and association studies from mid-latitude populations provide evidence for specific candidate locus-trait associations. We show how the performance of selected lines in field cages can validate traits under selection and how parallel clines in related species can validate the adaptive nature of the clines.

Cryoprotective dehydration – cold tolerance in permeable invertebrates

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Besides the two well-known cold hardiness strategies, freeze avoidance and freeze tolerance, a third, and fundamentally different, mechanism to survive freezing temperatures by dehydration exists in organisms with highly permeable integument. When soil water in their environment freezes, they dehydrate due to the difference in water vapour pressure between the supercooled body fluids and the surrounding ice. Dehydration continues until vapour pressure equilibrium between internal fluids and the surrounding ice is reached. Recent studies have shown that this cold hardiness mechanism is widespread in soil invertebrates (e.g. Collembola) and coincides with high cuticular permeability to water. In such animals, supercooling capacity is of little or no importance for winter survival. Dehydration triggers physiological changes in Collembola, such as accumulation of sugars and polyols, expression of heat shock proteins, and shifts in the phospholipid composition of membranes. These changes are likely to promote cold tolerance in a dehydrated state.

Parasitoid foraging behavior in a life history context

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In the 1980's a synthesis between Behavioral Ecology and Life History Theory was initiated by including life history parameters into state dependent optimal foraging models. To test these models, we collected empirical data showing that in the Australian parasitoid *Asobara persimilis* (Hymenoptera: Braconidae), genetic differences in life history traits correlate with differences in foraging behavior. *A. persimilis* occurs along the entire Australian east coast. We studied life span, patch time allocation and oviposition behavior of parasitoid populations from the Adelaide, Melbourne, Sydney and Brisbane regions. Life span increased along a gradient from Adelaide to Brisbane. Correlated to this, patch residence times and oviposition activity decreased along the same gradient. These results are in accordance with foraging models predicting a negative correlation between life expectancy of time-limited foragers and the degree to which they should exploit patches of a resource with a clumped distribution.

Exploring winter energy consumption as a limitation to the southern range limit of the goldenrod gall fly (*Eurosta solidaginis*) in late goldenrod (*Solidago gigantea*)

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The goldenrod gall fly, *Eurosta solidaginis*, is a gall-forming insect that has diverged into two host races on its hosts *Solidago gigantea* and *S. altissima.* These host races show appreciable reproductive isolation and genetic differentiation. Both host plants range from Ontario to Florida in eastern North America. Flies of the *altissima* host race can be found virtually everywhere its host plant is present, but *gigantea* flies are only present in the northern tier of its host's range. We performed a series of experiments to identify factors restricting the *gigantea* host race to northern locales. Correlation analysis suggests that the southern limit of *gigantea* flies is constrained by warm winters, likely because warmth increases energy consumption. *Gigantea* flies are smaller and less fecund because *S. gigantea* senesces earlier (thus ceasing feeding earlier in the autumn) than does *S. altissima.* Being smaller, the *gigantea* host race has less energy to support metabolism during winter and egg production the following spring. We will present our ongoing efforts to produce a model of winter energy use for goldenrod gall flies across their range, and consequent range limitations. Our work will stand as a model for predicting shifts in geographic distributions with global climate change.

Sources of variation in costs of plant resistance to herbivores

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A meta-analysis of 70 studies assessing the relationship between plant resistance and fitness has been conducted to examine the importance of several sources of variation in the fitness cots of resistance. The greatest costs were observed under uncontrolled environmental conditions in the field, at high levels of nutrient availability, and for defenses associated with alkaloids and phenolics. The prevalence of costs in the field and their persistence in the presence of herbivores both imply that fitness costs may arise not only due to trade-offs in the allocation of common limited resources between defense, growth and reproduction within an individual plant (direct allocation costs), but also through interactions between plants and external factors in their environment (ecological and opportunity costs). Cumulative meta-analysis revealed the evidence of publication and citation bias in studies on costs of plant resistance. The early studies conducted in 1975-1990 reported significant negative correlations between defense and fitness measures. However, after publication of several theoretical studies that have provided theoretical justification for the absence of costs, the magnitude of the reported costs decreased and became non-significant in the early 1990s. Furthermore, studies reporting significant costs of resistance received more citations than those, which reported no evidence of costs.

The evolution of egg retention and sex allocation: a study of facultatively ovoviviparous thrips

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Idolothripine thrips provide an unparalleled opportunity for investigating the evolution of reproduction. In many species, females are facultatively ovoviviparous/viviparous, and individuals choose among egg laying, live birth or both. As thrips are haplodiploid, females have some control over the sex of their offspring. We present our preliminary phylogeny and contrast work for the subfamily, and a case study of the Japanese idolothripine, *Bactrothrips brevitubus*. In this species, both sexes are produced by oviparity but only males are produced by ovoviviparity, so that sex allocation and reproductive mode act in tandem. Local resource competition between females for food and egg-laying sites may select for ovoviviparity, as females can retain their eggs until a better rearing site is located. Predatory mites could also select for ovoviviparity, as they consume the eggs but are non-fatal to larvae and adults. Population-wide sex ratios are generally unbiased, but our DNA and life history data indicate that the sex ratios are not Fisherian and are split according to the dominant reproductive mode in a patch. We consider how females tradeoff the best reproductive mode and sex allocation strategies. More broadly, we consider how egg retention and sex allocation may have coevolved in the subfamily.

Rapid cold-hardening: mechanisms and ecological significance

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More than 15 years ago we reported a rapid cold-hardening (RCH) response that protects various species of insects and related taxa against cold shock (non-freezing) injury. This swift acclimatory response to a brief exposure at a moderately low temperature allows the insect to survive a subsequent cold shock that would otherwise be lethal. Although little is known about the physiological mechanisms underpinning this response, recent evidence suggests that modifications of membrane fluidity and stress proteins play a role. RCH can also occur in isolated cells directly exposed to low temperature. Originally, we hypothesized that the RCH response allowed insects to enhance "instantaneously" their cold tolerance in a thermally variable environment. Recently, we obtained strong support for this hypothesis by demonstrating that RCH occurs under ecologically relevant cooling rates and diurnal thermoperiods. Furthermore, the rapid acclimatory response is not restricted to cryoprotection, but also functions to enhance organismal function at multiple levels, including courtship and mating, in response to environmental cooling at moderate to high temperatures. Thus, it is clear that RCH has broad ecological significance because it allows insects to continuously fine-tune organismal response to match even modest changes in environmental temperature that occur throughout the year.

Costs and benefits of resistance and tolerance to herbivores: the importance of gene x environment effects

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Trembling aspen (*Populus tremuloides*) is the most widespread tree species in North America and the most genetically variable plant species known to science. Extraordinary genetic variation of this clonal species extends to traits such as rates of growth and levels of chemical (salicylates and tannins) defense. Given that aspen is routinely subject to expansive defoliation events (e.g., large aspen tortrix, forest tent caterpillar) the question arises as to why defense polymorphisms persist in natural populations, i.e., why do not all genotypes express maximum levels of defense? The emerging answer involves both environmental and ontogenetic influences on the expression of chemical defense. For young aspen trees, high commitment to chemical defense incurs a significant cost in terms of growth in resource-poor, but not resource-rich, environments. Thus, in the absence of herbivory, highly defended trees are disadvantaged because slow growth may limit competitive ability. As trees mature, salicylate-based defenses tend to decline, extensive root systems develop, and trees appear to shift their defensive strategy to tolerance. Tolerance itself is also genetically variable and influenced by resource availability.

Sexual isolation in North American fruit flies: reinforcement, reproductive character displacement and the role of courtship song

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Differences in the courtship song of *Drosophila* males, produced by wing vibrations, have been implicated in sexual isolation between several species and races through female discrimination. The range of *Drosophila persimilis,* California, Oregon and Washington State, is completely contained within that of its sibling species *D. pseudoobscura* which extends as far east as Texas. The species are morphologically identical but sexually isolated. Although male courtship appears to be indiscriminate, females of these species discriminate against mating with heterospecific males. Females from areas where they co-occur discriminate more than those collected from allopatric locations. This has been suggested to be an example of speciation by reinforcement. Despite identifying species differences in both major song parameters, interpulse interval (IPI) and intrapulse frequency, previous work has suggested that these song differences are not responsible for sexual isolation. In this study playback experiments of synthetic *D. pseudoobscura* and *D. persimilis* song demonstrate clearly that females do indeed discriminate between conspecific and heterospecific courtship song. Reinforcement may be as a result of a narrowing in female preferences in sympatry or of reproductive character displacement (RCD) in a male courtship trait. The courtship songs of *D. pseudoobscura* from both sympatric and allopatric locations are examined for evidence of RCD.

Adaptive responses by Australian burrowing cockroaches to fluctuating Neogene and Pleistocene environmental conditions: evolution in parallel and reverse?

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Over the last ~15 million years, many areas in Australia once covered by rainforest have been transformed into zones of savanna, shrubland and sclerophyll forest, primarily due to long periods of arid climatic conditions. Understanding how the ancient ancestors of Australia's unique extant fauna responded to these changes is of singular interest. We have recently provided phylogenetic evidence that Australian-native soil-burrowing cockroaches evolved from wood-feeding ancestors following an invasion of the latter from Asia into Australia. The transition from wood to soil burrowing was most likely a result of aridification, which would have led to the disappearance in many areas of wood suitable for burrowing. We have extended our study to include all known soil-burrowing cockroach species, as well as most Australian burrowing cockroach species. Phylogenetic analyses based on mitochondrial COII data suggest that the soil-burrowing habit may have evolved on more than one occasion. An alternative explanation is that it evolved only once, but has been followed by 're-evolution' of the wood-burrowing habit. Approximate times of divergence between key taxa will be estimated using molecular dating methods, allowing a preliminary examination of whether these transitions occurred at similar or different times.

Does predation maintain plasticity in *Bicyclus anynana*?

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The butterfly *Bicyclus anynana* exhibits phenotypic plasticity involving the wet season phenotype which possesses marginal eyespots on the ventral surface of the wings and the dry season form that lacks these eyespots. We examined the adaptive value of phenotypic plasticity of *B. anynana* in relation to the defence mechanisms of crypsis and deflection. We assessed the visibility differences between spotless and spotted butterflies against backgrounds of brown (y season) or green (wet season) leaves. Spotless butterflies were highly cryptic and less predated by adult bird predators than spotted ones when presented against brown leaf litter. The advantage of crypsis disappeared in the wet season habitat as both forms were equally visible. In later experiments, naïve birds presented with resting butterflies in the wet season habitat tended to learn more rapidly to capture spotless butterflies, suggesting a slight selective advantage of possessing eyespots. Marginal eyespots increased significantly the escape probability of butterflies that were attacked by naïve birds compared to adult birds, although there were no differences in prey capture success within naïve predators. Our results show that natural selection acts against eyespots in the dry season favouring crypsis whereas in the wet season it may favour eyespots as deflective patterns.

The peppered moth: decline of a Darwinian disciple

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The evolution of melanism in the peppered moth, following the industrial revolution in Britain, is the most cited example of an observed heritable change that resulted from Darwinian evolution. The reason for this is that the change was visually obvious, was observed, and could be explained by Darwin's central mechanism of evolution - natural selection. Further, the reason that the black form was fitter than the pale form was easy to understand: against lichen denuded and smoke blackened bark, the black moths were better camouflaged, and so were less often found and eaten by birds. Recently creationists have seized upon design flaws in Bernard Kettlewell's classical experiments in the 1950s. They claim that these flaws, and more recent allegations of fraud aimed at Kettlewell and a conspiracy of silence among evolutionary biologists, particularly in Britain, fatally undermine this cornerstone of Darwinian evolution, and therefore the basic theory of evolution by natural selection. Many schools in the U.S.A. have dropped the peppered moth case from biology teaching, and pressure is growing elsewhere. Most biologists who have worked on the peppered moth take the view that although Kettlewell's experiments did contain design flaws, these flaws would only have affected the quantitative accuracy of his tests; not the qualitative interpretation that differential bird predation has played a major role in the evolution of melanism in the peppered moth. The question I will address is whether melanism in the peppered moth should still be regarded as an example, par excellance, of Darwinian evolution in action.

Origin and evolution of seasonal adaptations: ground crickets in tropical, subtropical and temperate climatic conditions

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The ground cricket species complex, *Dianemobius nigrofasciatus* and *D. fascipes*, is probably of tropical origin and widely distributed from tropical to temperate regions in East Asia. It might be surmised that as peripheral populations dispersed northward, they encountered gradual changes in climatic conditions, evolved locally different types of seasonal life cycle, and egg diapause became the core of adaptation to overwintering in the northern climate. I pursued the origin of egg diapause and the associated patterns of nymphal development in local populations at low (tropical and subtropical) latitudes and found dual responses (egg diapause and delayed nymphal development) to short days in some of them. In other tropical populations a few eggs showed delayed hatching. The occurrence of such eggs might represent 'life-cycle polymorphism' to cope with unpredictable risks after hatching and might be the source of evolution for egg diapause and overwintering in the north.

Ontogenetics of QTL: the genetic architecture of trichome density over time in *Arabidopsis thaliana*

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Although much is known about the molecular genetic basis of trichome development in *Arabidopsis thaliana*, less is known about the underlying genetic basis of continuous variation in a trait known to be of adaptive importance: trichome density. The density of leaf trichomes is known to be a major determinant of herbivore damage in natural populations of *A. thaliana* and herbivores are a significant selective force on genetic variation for trichome density. A number of developmental changes occur during ontogeny in *A. thaliana*, including changes in trichome density. I used multiple interval mapping (MIM) analysis to identify QTL responsible for trichome density on both juvenile leaves and adult leaves in replicate, independent trials and asked whether those QTL changed with ontogeny. In both juvenile and adult leaves, I detected a single major QTL on chromosome 2 that explained much of the genetic variance. Although additional QTL were detected, there were no consistent differences in the genetic architecture of trichome density measured on juvenile and adult leaves. The finding of a single QTL of major effect for a trait of known adaptive importance suggests that genes of major effect may play an important role in adaptation.

Regulation of a plant allelochemical-inducible detoxification gene via the aryl hydrocarbon response cascade

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The promoter of *CYP6B1*, a cytochrome P450 monooxygenase (P450s) that metabolizes furanocoumarins (i.e. xanthotoxin) found in the specialized diet of the swallowtail caterpillar *Papilio polyxenes*, utilizes several elements for basal and xanthotoxin-inducible expression, including the xenobiotic response elements to xanthotoxin (XRE-Xan) and aryl hydrocarbons (XRE-AhR). In mammalian systems, XRE-AhR mediates induction of *CYP1A1* by aryl hydrocarbons via the aryl hydrocarbon receptor (AhR) and its heterodimeric partner, ARNT. To determine if AhR-like and ARNT-like proteins from *Drosophila melanogaster*, spineless-aristapedia (Ss) and tango (Tgo), are capable of activating *CYP6B1* transcription via its XRE-AhR element, Ss and Tgo proteins were coexpressed with the wildtype *CYP6B1* promoter:CAT construct in an Sf9 cell transfection system. These studies indicate that basal and xanthotoxin-inducible expression are significantly increased in the presence of Ss and that xanthotoxin-inducible expression is increased in the presence of Tgo. Coexpression of Ss/Tgo with the *CYP6B1* promoter constructs containing mutations in the XRE-Xan or XRE-AhR elements indicate that mutations in the XRE-AhR element reduce basal and xanthotoxin inducible expression but retain low levels of activation in the presence of Ss/Tgo. These results suggest that the *CYP6B1* promoter employs conserved elements of the aryl hydrocarbon regulatory cascade for basal and xanthotoxin-inducible expression that are shared between vertebrates and invertebrates (XRE-AhR).

"*Drosophila melanogaster* gene expression in response to a virulent endosymbiont "

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Wolbachia pipientis is an obligate intracellular bacterium present in a wide range of insect species. While the bacterium is known for its ability to manipulate host reproductive biology, most strains cause little direct harm to host tissues. One exception is a strain called "popcorn" that infects *Drosophila melanogaster.* As the fly ages the bacterium replicates to large numbers, leading to rupture of host cells and shortened insect lifespan. This over-replication is aberrant in comparison with other *Wolbachia* infections whose densities remain stable or decline with insect age. Previous work indicates that *Wolbachia* infections do not induce or actively suppress an innate immune response in *Drosophila melanogaster.* Because the popcorn infection leads to cell rupture we predicted that this strain would likely induce host defenses. Using a microarray approach we have examined the effect of popcorn infection on host gene expression as the host ages. Overall, gene expression decreases with age in response to popcorn infection. The bacterial specific response resulting in antimicrobial peptide production is not activated. The most common class of upregulated genes, however, were serine proteases; including elastases and trypsins. Serpins, the regulators of these proteases are also highly expressed, as are other genes encoding general defense response proteins.

Evolutionary dynamics of *w*Au-like *Wolbachia* strains in Neotropical *Drosophila* species

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We have screened 29 American strains of the Neotropical species *Drosophila willistoni* for the infection status with the -proteobacterium *Wolbachia wsp* PCR diagnostics and Southerns blots. Although *Wolbachia* is absent in older strains of *D. willistoni* collected before the 1970s more recent samples derived from natural populations all over the American continent are universally infected with the A-group *Wolbachia* strain named *w*Wil. *w*Wil of *D. willistoni* is identical to *w*Au of *D. simulans*. Like *w*Au in *D. simulans* *w*Wil is not inducing cytoplasmatic incompatibility (CI) in *D. willistoni*. In contrast to *w*Au a strain infecting both germline and somatic tissues *w*Wil is exclusively targeting the primordial germline cells of *D. willistoni* embryos. This tissue tropism of *w*Wil and its temporal and spatial biogeographically pattern strongly support (*i*) a long-term evolutionary relationship between *w*Wil and its host species *D. willistoni* in some isolates; (*ii*) its modern day infective spreading throughout *D. willistoni* American populations, and (*iii*) our assumption that this Neotropical *Drosophila* species can be regarded as the most likely donor source for a recent *Wolbachia* infection of immigrating *D. simulans* with the strain *w*Au. Furthermore new *Wolbachia* strains derived from Neotropical *saltans* group species do harbor polymorphic *w*Au-like *Wolbachias*.

Mechanism and evolution of caste polyphenism in social insects

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Phenotypes of organisms are not determined completely genetically, but vary according to environmental factors (phenotypic plasticity). Some organisms express several discrete adaptive phenotypes (polyphenism). Social insects possess a few types of individuals (castes) in their colonies, to which tasks are allocated. Here, I introduce our case studies related to caste polyphenism in ants and termites. We focus on the developmental mechanisms of cast-specific characters, such as alate wings or soldier mandibles. In ants, the developmental fate of caste is determined in the early stage of postembryonic development, but apoptotic degeneration occurs in the wing primordia of future workers. As apoptotic wing degeneration was observed in two phylogenetically distant groups of ants, this phenomenon is suggested to be common in many ant species. On the other hand, all termite species possess distinct sterile soldiers, which have specific morphology suitable for defense. We isolated genes related to soldier differentiation and analyzed the expression profiles of those genes, in order to understand the mechanism of caste differentiation and the link between molecular and social evolution. I will compare these studies, in terms of the alteration of body plan in response to environmental signals, and discuss the evolutionary process of the interaction between ontogeny and environment.

Possible reproductive isolation as an indirect consequence of adaptation to different environments: pleiotropic effect of clock genes

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Clock genes pleiotropically controlling development time, circadian rhythm and time of mating can cause an allochronic reproductive isolation in a model insect, the melon fly *Bactrocera cucurbitae*, which mate once a day at dusk. A series of experiments including artificial selection and behavioural analyses showed that flies with shorter developmental and circadian periods (ca 22 h of locomotor activity rhythm) mated 5 h earlier in the day than those with longer periods (ca 30 h). Mate choice tests demonstrated significant pre-mating isolation between populations with short and long circadian periods. Pre-mating isolation did not occur when the mating time was synchronized between the two populations by photoperiodic controls, indicating that reproductive isolation is due to variations in the time of mating and not any unidentified ethological difference between the two populations. The fluctuation of *per* gene mRNA expression under DD is closely correlated to the locomotor rhythm in each population with short and long periods in *B. cucurbitae.* The results suggest that a clock gene might be involved in the allochronic pre-mating isolation. I discuss allochronic reproductive isolations as an indirect consequence of adaptation to different environments in view of a pleiotropic effect of clock gene controlling time-related traits.

The development and evolution of novel phenotypes: a case study on beetle horns

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How do ecological, developmental, and genetic mechanisms interact in the origin and diversification of novel phenotypes? This question is at the center of my research on the evolution and development of beetle horns, a morphological novelty that upon invention has undergone dramatic diversification on a variety of levels. Horn expression is modulated in the context of alternative male morphologies, sexual dimorphisms, between-population differences in allometric scaling, and between-species differences in the number and location of horn development. Combining morphometric, candidate-gene, and ecological approaches my research aims to integrate a genetic and developmental understanding of phenotype expression into the behavioral and ecological contexts within which different phenotypes function. In my talk I will first focus on how a developmental genetic understanding of horn expression can inform us about the origins of non-linear scaling relationships, sexual dimorphisms, and the parallel gain and loss of horns in similar and different body regions. I will then explore which ecological and social conditions contribute to and guide evolutionary changes in beetle horn development. Beetle horns and horned beetles emerge as an outstanding model system for studying the interplay between ecological and developmental mechanisms is the origins of phenotypic diversity.

Attempts to search for host genes affected by intracellular symbiotes in their expression level: transcriptome analyses with silkworm cDNA microarray

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Arthropods harbor intracellular bacteria, such as *Wolbachia* and *Cardinium* (CFB [*Cytophaga-Flavobacterium-Bacteroides* phylum] bacteria). These facultative bacteria cause reproductive alteration in their hosts such as thelytokous parthenogenesis, male killing, feminization and cytoplasmic incompatibility. As a part of the attempt to investigate the effects of these bacteria on hosts, we examined gene expression in bacteria-infected and non-infected cells. *Wolbachia* of the small brown planthopper *Laodelphax striatellus* and Is-CFB (CFB bacteria isolated from *Ixodes scapularis*) were cultivated in silkworm cell lines (BmN4 and NIAS-Bm-aff3). A cDNA microarray, which contains about 6,000 cDNA clones of the silkworm, was used to identify the genes that were affected in the expression level by the infection of *Wolbachia* or Is-CFB. No clearly up-regulated genes were found by *Wolbachia* infection, but one transcript was down-regulated. Is-CFB infection raised expression level of a few genes.

Female genitalia and sexual selection: a morphological study of cockroach genitalia

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The male genitalia of Hexapoda are often complex and species-specific in form. Several hypotheses have been proposed to explain this. This study tests three hypotheses; Lock-and-Key, Female Choice and Male-Female Conflict by studying the genitalia of four species of cockroaches. The interacting parts of cockroach genitalia are on the morphological outer side of the body and are partly sclerotised in both sexes. Furthermore, cockroach male genitalia are highly asymmetric and the direction of asymmetry has been reversed several times. These traits make cockroach genitalia easily accessible, morphologically variable, and thus highly suitable for the study of genitalic evolution.

Contrary to "Lock-and-Key," the asymmetry of the male genitalia is not matched by asymmetry in the female genitalia. As predicted by "Female Choice", mechanoreceptors are found in the areas of genitalic interaction, but a reversal of male genitalia asymmetry does not influence their placement. There is quite strong evidence against "Male-Female Conflict" as the female genitalia facilitate gripping by the male through a ledge or other holding points. However, in water-striders "Male-Female Conflict" is well documented. This suggests that none of the proposed hypotheses can explain genitalic evolution in the Hexapoda by itself, thus the ultimate explanation must be something different.

Genetic diversity and oviposition host preference of *Bemisia tabaci* (Gennadius) (Homoptera: Aleyrodidae) associated with four crops in Ghana

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The genetic and behavioural diversity of populations of the polyphagous virus vector, *B. tabaci* infesting cassava, tomato, garden egg and okra in Ghana were assessed. Seventy-six individuals collected from four hosts in three locations were screened by RAPD-PCR, using five primers (OPA 02, OPB 08, OPC 05, OPD 16 and OPI 16). Cluster analysis using the Dice coefficient revealed two genetic clusters at the 45% level of similarity associated with cassava and okra. Crop association contributed more to the observed variability than geographical isolation. Two-choice oviposition preference bioassay was done in a petri-dish cage. Cassava whiteflies significantly ($p < 0.01$) preferred cassava to any other host, but preferred garden-egg or tomato in the absence of cassava. Tomato populations significantly ($p < 0.05$) preferred tomato and okra, while okra whiteflies did not oviposit on cassava. These results corroborate the existence of two distinct biotypes: the cassava biotype and the polyphagous biotype. Tomato, garden egg and eggplant are potential common hosts for the two and may be reservoir hosts for under intensive systems or in rotation programmes. It is suggested that whitefly management on the main hosts should accommodate the possible spill of resistance from these hosts into cassava and vegetable crops.

Molecular characterization of different biological forms of *Anopheles stephensi* Liston in Iran

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Malaria is still one of the main health problems in south and southeast provinces of Iran. Mosquitoes of *Anopheles stephensi* are the most important malaria vector in these regions. Based on egg ridges three biological forms of type, intermediate, and *mysorensis* has been reported within this species which shows different vector capacity and competence. Here we study the sequence variation of 1500 bp of mtDNA cytochrome oxidase subunit I (COI) of these biological forms to find molecular marker for diagnostic purposes. Different biological forms were collected from different parts of the country and were subjected to PCR-RFLP or PCR sequencing assays. Results of PCR-RFLP showed no polymorphism between different biological forms or populations. This was confirmed by comparing 1400 bp sequences of COI and COII regions which showed a high similarity between forms. However there were two haplotypes between populations of *mysorensis* form. These findings are in agreement with previous hybridization, cytological, and molecular studies stating a great gene flow between populations of *An. stephensi.*

mtDNA PCR-RFLP and PCR sequencing revealed significant genetic variation among *Anopheles superpictus* populations in Baluchistan, Iran

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Mosquitoes of *Anopheles superpictus* are one of the main malaria vectors in Iran and present in most parts of the country. This taxa play different rules in malaria transmission in different areas of its geographical distribution. This study has been conducted to investigate rate of genetic variation between different populations of *An. Superpictus* in Baluchistan, the most important malarious region in Iran. A portion of about 1512 bp long of mitochondrial DNA cytochrome oxidase subunits I and II were amplified and were assayed by either RFLP or direct sequencing. Analysis RFLP of COI-II and sequences of 750 bp of 3′ end of COI gene showed that there are two main groups within specimens. Rate of sequence variations were up to 5 percent which is far exceeding than inter-population or -species variation. It is suggested that presumably this species is a group species. The results of this study which is the first molecular study on *An. superpictus*, needs to be confirmed by further molecular, cytological, and morphological studies and checked with available epidemiological data to reveal the relationship between these haplotypes and malaria transmission in each locality.

Micromalthus debilis **LeConte and its varied symbionts**

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The wood-boring telephone pole beetle, *Micromalthus debilis*, is the only member of the primitive family Micromalthidae. It has fascinated entomologists for years because of its unique life history features such as haplodiploidy, parthenogenesis, paedogenesis, and matriphagy. Many aspects of its biology still remain a mystery due to complications in rearing and manipulating the fragile individuals and broods. In spite of the scarce experimental data available, speculations on the evolutionary biology of this insect have been plentiful; for example, obligate matriphagy by males has been linked to the presence of a non-functional nutritional endosymbiont suggesting that *Micromalthus* is a case of Hamilton's hypothesis about the evolution of haplodiploidy, and the high cost of producing males has created a selective advantage for cyclic thelytoky. Using microscopical and molecular tools we are screening the symbiotic load of the different larvae and adults of *Micromalthus*. Several symbionts have been identified so far, including renowned reproductive parasites. We discuss the life history traits and evolutionary biology of this beetle in relation with our results and the comprehensive early literature.

Rapid cold hardening increases aphid survival at low temperatures.

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Most studies on insect cold hardiness have focused on seasonal adaptations occurring within the overwintering stage of an insect's life cycle, triggered by decreasing temperature and photoperiod from summer through autumn to winter. Due to their short generation times, an aphid that enters winter is unlikely to be the same individual that survives until spring. For this reason, although aphids are able to cold harden when reared at lower temperatures (10° and 5°C), the ability to rapidly cold harden may be relatively more important than in other longer-lived overwintering insects, where seasonally-induced cold hardening is the dominant response. This study assesses the effects of sub-zero temperatures on the mortality of nymphs and adult *S. avenae*, provides evidence of rapid cold hardening in *S. avenae* and investigates the ecological costs of rapid cold hardening in terms of effects on development, longevity and fecundity. The hypothesis that rapid cold hardening can be induced during the cooling phase of natural diurnal temperature cycles is supported. Rapid cold hardening increases survival and extends the lower lethal temperature, allowing aphids to acquire maximum cold hardiness when it is most needed.

Using geometric morphometrics to investigate wing dimorphism in males and females of hymenoptera – a case study based on the genus *Tachysphex* (Sphecidae: Larrinae)

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Traditional morphometrics are currently complimented by an additional statistical method, namely geometric morphometrics which is based on the quantification of shape using *x,y* coordinates. It is characterized by the obtaining of an exact geometric description of the morphology, and the ease to visually determine which landmarks are responsible for differences/similarities. The amount of variation between the forewings of males and females of the genus *Tachyshpex* (Hymenoptera) is investigated here. The research investigates the extent of morphological similarities/differences between males and females of the same species, in order to make recommendations regarding inclusion of both sexes in a systematic or geometric morphometrics study. Mantel statistics revealed a good correlation between the male and female dataset, and although visual analysis of the RWA plots of the two sexes indicated significant correlations between sexes of the same species, differences in the position of the species clusters suggests that sexual dimorphism in wings are present. Results from the thin-plate spline analysis of the mean shape of the male and female dataset revealed which landmarks are responsible for the differences. It is concluded that, in an analysis of members of a genus, only one of the sexes be used in the analysis, as small-scale differences may influence the results.

Glue protein in *Drosophila*: protein with an obscure function?

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Perusal of literature from the first description of larval salivary gland secretory protein (Fraenkel & Brookes, 1953) in *Drosophila* namely "Glue Protein", to the recently published article suggests that the glue 'helps' pupa to 'fix' to a substratum. Several lines of genetic and biochemical investigations on this tissue specific and stage specific protein in different species/subspecies of *Drosophila nasuta* subgroup has revealed that (a) abundant glycosylated protein is synthesized (b) most protein fractions are synthesized by co-dominant X-chromosomal gene/s (c) SDS-PAGE mobility differences of major glue fractions probably reflect differences in amino acid composition (d) there is exuberant SDS-PAGE pattern polymorphism (e) there is heteroselection of X-chromosomal glue gene/s in females in the natural populations of *D. nasuta nasuta* and *D. sulfurigaster neonasuta*. Morphometric, quantitative and behavioural studies involving several *Drosophila* species revealed absence of correlation between larval/pupal/gland size and the quantity of secretions. Species producing abundant glue prefer to pupate in the medium, those producing lesser quantities pupate on solid surface, while those producing negligible quantities, pupate in cotton plugs. These results suggest that the larval salivary gland secretions in *Drosophila* have other functions, in addition/rather than to help the pupa to 'fix' to a solid surface.

The evolution of sexually-selected traits in the sand cricket, *Gryllus firmus*

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Sexually-selected traits typically consist of components of morphology, physiology and behavior. This is particularly evident in Orthoptera where males call to attract females. Male call song is determined by the morphology of the sound-generating structure, physiological costs of calling and female behavior. In this talk I explore the evolution of these components in the cricket, *Gryllus firmus.* I discuss the genetic basis of the morphological components that generate the song, the song components themselves and the underlying physiology. I show that female choice is determined by both presence/absence of song and by song components. Because of the physiological costs of calling, macropterous males call substantially less than macropterous males and attract fewer females. Thus sexual selection can act not only on the components of the traits involved in female attraction but also on traits necessary for dispersal and hence can potentially have far-reaching affects on fitness. A common garden experiment using three geographically separate populations shows that the components underlying female choice vary among populations. Further, comparison of female choice in a population maintained in the lab for twenty years shows that this choice is stable over time, suggesting that it is maintained due to pleiotropy and not linkage disequilibrium.

The evolution of genetic architecture in response to environmental change

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Environments are highly variable in both space and time. Insects, being ectothermic are particularly sensitive to changes in their thermal environment. Evolutionary response to changes in the thermal environment are contingent on the presence of available additive genetic variation and the direct effect of the environment on the expression of this variation. I discuss two methods of investigating these factors: the experimental manipulation of temperature and the analysis, using a common garden experiment, of genetic variation along clinal gradients. For the experimental work I describe work with the cricket *Gryllus firmus*, showing that changes in temperature can have highly significant effects on phenotypic expression but relatively little on the genetic variance-covariance matrix. However, there are differences between wing morphs suggesting that these morphs show differential canalization responses to temperature change. For the work on geographic variation I describe common garden experiments conducted on two species of grasshopper (*Melanoplus* spp). These experiments show that the clinal variation in phenotypic traits is matched by clinal variation in genetic variation and that the differences among populations is concordant with previous phylogenetic analysis.

Reinvestigation of the mechanism of the female–biased sex ratio in *Wolbachia*–infected *Ostrinia furnacalis* (Lepidoptera: Crambidae)

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Wolbachia-infected females of *Ostrinia furnacalis* and *O. scapulalis* produce all-female offspring. The all-female production was interpreted as feminization of genetic males, since the antibiotic elimination of *Wolbachia* resulted in all-male offspring. However, in *O. scapulalis*, observation of the sex chromatin suggested that male-killing is the cause of the all-female production and that female-specific death is the cause of the all-male production after *Wolbachia* elimination. In the present study, we observed chromosomes of infected and cured insects of *O. furnacalis* to reinvestigate mechanism of the all-female production in this species. Infected female adults were found to be genetic females based on the presence of the female-specific sex chromatin. Both genetic males and females were included in the hatched larvae in the broods derived from infected females. Likewise, both genetic males and females were found in the hatched larvae in most broods derived from cured females. In contrast, the last instar larvae derived from infected females were in the all-female condition, and those derived from cured females were in the all-male condition. These results suggested the occurrence of opposite sex specific lethality in *Wolbachia*-infected and cured females of *O. furnacalis*, as suggested in *O. scapulalis*.

Paropsis atomaria (Olivier) (Coleoptera: Chrysomelidae): The significance of genetic and ecological diversity in a wide-ranging insect species

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The *Paropsis atomaria* complex currently consists of three described species, *P. atomaria*, *P. deboeri* and *P. elytrura*. While the latter two species have restricted distributions (to Tasmania and South-west Western Australia, respectively), *P .atomaria* is widely distributed in temperate, sub-tropical and tropical eastern Australia. The species are all *Eucalyptus* herbivores and are morphologically and behaviourally similar. This paper reports on a detailed investigation of both inter- and intra-specific relationships within the complex. Using molecular, morphological and ecological approaches the specific status of the three named taxa, as well as population level differentiation within the widely distributed *P. atomaria,* is being investigated. Special attention is paid to *P. atomaria* as it is an important forestry pest and there is potential that elucidation of population characteristics may reveal genetically distinct populations, possibly cryptic species, within the taxon. Assessment of the mitochondrial genome (COI, 550 b.p.) revealed approximately 9% differentiation between mainland *P. atomaria* and *P. deboeri*, whilst demonstrating a maximum of approximately 1.5% differentiation within *P. atomaria.* This supports current morphological classification. Such a multi-disciplinary, holistic approach will ultimately provide a robust foundation for the understanding of species boundaries and population dynamics in this economically important complex.

Adaptation to environmental gradients.

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Latitudinal clines are widespread and provide a natural framework within which to examine natural selection. Repeated clines in different geographic areas or in related taxa strongly implicate the occurrence of natural selection. Clinal variation at the phenotypic level therefore provides an opportunity to examine the way in which genetic variation may also be influenced by natural selection. Despite the widespread occurrence of latitudinal clines, there are few empirical data that address the question of how quantitative genetic variation varies along such clines. We have combined comparisons of clinal populations of *Drosophila serrata* collected from the eastern coast of Australia with laboratory natural selection experiments to examine the genetic basis of adaptation to environmental gradients. We show that clinal selection under natural conditions does result in changes in both the additive and nonadditive genetic variances. Furthermore, we show that adaptation to clinal environments may involve a number of distinct genetic responses along the length of the cline, the complexity of which may not be fully revealed by focussing primarily on populations at the ends of the cline.

Metabolism of the sub-Antarctic caterpillar *Pringleophaga marioni* during cooling, freezing and thawing

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Although general models of the processes involved in insect survival of freezing exist, there have been few studies directly investigating physiological processes during cooling, freezing and thawing, without which those models remain hypothetical. Here we use open-flow respirometry to investigate metabolism of the freeze-tolerant sub-Antarctic caterpillar *Pringleophaga marioni* Viette (Lepidoptera: Tineidae) during cooling, freezing and thawing, and to compare animals exposed to non-lethal (-5.8 °C), and lethal (-6.0 °C, after which caterpillars are moribund for several days; and -18 °C, after which caterpillars are completely unresponsive) freezing stress. We found a large decrease in metabolic rate (that is not associated with freezing) at -0.6 ± 0.1 °C, and calculated a Q_{10} of 2.14×10^3 at this breakpoint. This breakpoint is coincident with the critical thermal minimum, and is hypothesised to be a metabolic manifestation of the latter. Caterpillars froze at -4.6 ± 0.1 °C, and had non-zero metabolic rates when frozen. Post-thaw, metabolic rates were lower than pre-freezing measurements. Post-thaw metabolic rates did not differ between temperatures that did and did not kill the caterpillars, which suggests that mortality may be a result of a breakdown in processes at the organismal, rather than cellular, level of organization.

Evolutionary implications of infection with parthenogenesis inducing *Wolbachia*

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The presence of Parthenogenesis Inducing (PI) *Wolbachia* infected individuals can have a strong impact on both the infected individuals as well as on the uninfected individuals present in a population. Two situations exist, the infection remains at some equilibrium or the infection goes to fixation. In this presentation I will present a model showing that an equilibrium infection frequency can only exist at a low level of infection if there are some factors present in the population that counteract the *Wolbachia* by modifying its expression /transmission (suppressor), or when other genetic elements are present that exploit the female biased sex ratio caused by the infection. If populations have gone to fixation for the infection with PI *Wolbachia* this generally is accompanied by a change in the females mating behavior and/or fertilization behavior. These changes are such that females from infected lines are unable to successfully mate and/or fertilize their eggs, whereas males derived from these same infected lines (through antibiotic treatment) are still functional and capable of fathering offspring.

Does environmental stress increase genetic structure in the springtail *Orchesella cincta*?

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Environmental stress can alter the genetic composition of natural populations by directional selection towards tolerant genotypes, by causing bottlenecks and by changing gene flow to and from stressed locations. Especially when gene flow is low, environmental stress is expected to increase levels of genetic differentiation among populations. We examined the effect of environmental pollution on population genetic structure for the soil dwelling species *Orchesella cincta* by comparing Isolation by Distance relationships. First, population genetic structure of sixteen NW European populations was estimated using six polymorphic microsatellite loci. A significant Isolation By Distance relationship was revealed, indicating gene flow is relatively high in this small soil arthropod species. Secondly, comparisons between stressed and pristine populations versus pristine populations revealed no differences in Isolation by Distance relationships. Our results indicate environmental pollution does not play a significant role in shaping population genetic structures within this species, most likely due to its migration capacity. Gene flow might, by filling up reduced genetic variability in stressed populations, be an evolutionary force maintaining versatility of stressed populations.

"Effects of eugregarine parasites on adult longevity in the polymorphic damselfly *Mnais costalis* Selys"

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Males of *M. costalis* show a genetic polymorphism linked to mate-securing strategies, typically existing as orange-winged territorial fighters and clear-winged nonterritorial sneaks (the former are larger in size) (Tsubaki 2003). Orange-winged males have a higher daily mating rate, however, their reproductive life span and longevity is less than that of clear winged males. As a result, the estimated lifetime reproductive success is the same (Tsubaki *et al.*, 1997). In this paper I will compare the survival cost of eugregarine infection for the two morphs looking at the relationship between the abundance of mid-gut parasites and the survival of field-collected males in captivity. Orange-winged males suffered higher parasite infection and showed shorter longevity than clear-winged males when they were fed at lower levels than the natural feeding rate, or when they were deprived of food. In contrast, morph longevity did not differ when they were fed until satiation. The survival of damselflies was negatively correlated with the parasite abundance except when damselflies were fed until satiation. The results suggest that the abundance of eugregarine parasites exerts a substantial cost, which is associated with the maintenance of alternative mating strategies, together with the higher developmental cost and energy expenditure of the fighter morph.

An evolutionary view of the sex communication in *Melittobia* spp.

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Melittobia are ectoparasitoids of late larval and pupal stages of several species of solitary bees and wasps. Females locate their hosts on which they will lay their eggs, and will represent the only nutritional resource to the developing larvae. Sex dimorphism is remarkable and males will fight to death to eliminate competing males as they mate emerging females. Males remain in the confines of the host's nest they developed in, while females may disperse to locate new hosts. Mating occurs after an elaborate courtship behavior, in which a male produced sex pheromone is involved, and is supposed to play an important role in mate recognition. We will describe the differences in the male pheromone of 4 species of *Melittobia* and evaluate the attractiveness of females to interspecific males, and discuss the role of the male sex pheromone and the cuticular hydrocarbon composition of these 4 species in sex mate recognition. We will discuss the evolutionary process involved in the chemical communication of this parasitoid and provide supportive molecular data.

The sociobiology of plant defense

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"Social context" can inflate or diminish the evolutionary success of a resistance gene by making genetic variance in fitness density/frequency-dependent. Three models illustrate. First, a plant growth model shows that intraspecific competition increases the benefit of resistance when attack levels are high (resistant plants usurp resources that damaged susceptible plants need for recovery), but also increases the costs when attack is absent (undamaged susceptible plants outgrow resistant ones). Next, a game theory model shows that the fitness benefit of resistance can vary through the interaction of enemy behavior with local genotype frequency: susceptible genotypes can gain associational resistance when at low frequency if enemies can move freely to more palatable patches; however, if enemies are restricted to single patches, the susceptibles are eventually found. Finally, plants with high allocation to resistance flower late, causing assortative mating for resistance under low herbivory. Heavy attack retards susceptibles, bringing them into flowering synchrony with resistants. A population genetic model shows that assortative mating inflates genetic variance during zero-herbivory periods when costs exceed benefits, but not during high herbivory periods when benefits exceed costs. This causes asymmetric selection responses that prevent fixation of resistance even if average benefit exceeds average cost.

The role of the hedgehog pathway in polyembryonic development

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Copidosoma floridanum is a polyembryonic parasitoid wasp that produces over 1000 individuals from a single egg. This clonal proliferative process is remarkable for a number of reasons but primarily because this insect system has evolved clonal proliferation as an adaptive strategy; therefore, understanding how cellular proliferation can occur without causing disease in this organism will increase our understanding of how the cell cycle is regulated in animals and how it might be possible to mimic this natural system. The Hedgehog (HH) signal transduction cascade is a candidate gene pathway that may be regulating proliferation within the dividing cells. Among metazoan taxa, one function of HH is to regulate cellular proliferation during development. Within *Drosophila,* for instance, HH has been shown to control the proliferation and differentiation of somatic stem cells and progeny within the ovary. To determine whether HH is implicated in polyembryonic proliferation in *C. floridanum*, we cloned the C. floridanum HH gene. Ultimately, we are interested in determining the role the *C. floridanum* HH signal transduction pathway has in polyembryonic proliferation. We predict that in areas of active cellular proliferation, high levels of HH will be translated, while in areas with no proliferation, HH expression will be absent.

The genetic basis of divergent gene expression in ***Drosophila***

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Differences in gene expression play a central role in evolution. Both *cis*- and *trans*-regulatory changes contribute to divergent gene expression, but their respective contributions remain largely unknown. Using closely related *Drosophila* species, we examined *cis*- and *trans*-regulatory changes underlying interspecific expression differences. Functional *cis*-regulatory changes were detected by comparing the relative abundance of *D. melanogaster* and *D. simulans* transcripts in F_1 hybrids, whereas differences in *trans*-regulatory activity were inferred by comparing the ratio of allelic expression in hybrids to the ratio of gene expression between species. We found that interspecific expression differences were almost always accompanied by changes in *cis*-regulatory function. In addition, *trans*-regulatory divergence affected approximately half of the genes we examined. These data contrast with recent studies of intraspecific gene expression, which found that expression variation was caused primarily by differences in *trans*-regulation. Distinct patterns of *cis*- and *trans*-regulatory changes appear to underlie expression differences within and between species.

Phenotype matching in wild parsnips and parsnip webworms: costs and consequences.

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The coevolutionary consequences of present-day interactions between plants and their insect herbivores may vary among populations as envisioned in the Geographic Mosaic Model of Coevolution. The interaction between wild parsnip, *Pastinaca sativa*, and its principal herbivore, the parsnip webworm (*Depressaria pastinacella*), is mediated by the amounts of furanocoumarins produced in the plant and the ability of the insect to detoxify them. Within 20 midwestern U.S. populations, trait frequencies in parsnips frequently were matched by trait frequencies in webworms. Exceptions were found in populations of parsnips with high xanthotoxin chemotypes, which are difficult for webworms to utilize, and in regions where an alternative host, *Heracleum lanatum*, which has a different furanocoumarin profile from wild parsnip, is abundant. The maintenance of more susceptible genotypes of wild parsnip in all populations is presumably due to the high cost of furanocoumarin production, while mismatching in alternate host populations is likely due to the cost of adaptation by webworms.

Anisotropic index of refraction of the iridescent scales of *Morpho* butterflies

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The ground scales of the blue iridescent *Morpho* butterflies are responsible of the coloration of the wings. Such a scale has been modelized as a 2dimensions photonic crystal with two very different periodicities: a nanoscopic vertical one in each ridge (typically 100 nm) leading to interferential effects, and a microscopic horizontal one, the ridges themself (~1 µm apart) giving a strong diffraction effect. We have modelized the actual structure of the scale as a graded grating, each ridge being equivalent to a multilayered mirror of alternatively high index films (cuticle material) and low index film (air with low fraction of cuticle material). In an effective medium framework, these two films are then assimilated to continuous films characterized by their effective refraction index. These index are determined by fitting the hemispherical reflectivity. Knowing the composition of each film, via electron microscopy, one can then deduced the cuticular material index. The cuticular material is equivalent to a uniaxial crystal, with the extraordinary axis parallel to the ridge. The two refractive index curves intersect in the 450-500 nm range, *i.e.* in the blue part of the spectra. The calculated value is in good agreement with the values usually found by liquid index technique.

The effects of dopamine receptor agonist and antagonists on the secretory rate of cockroach (*Periplaneta americana*) salivary glands

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The acinar salivary glands of the cockroach, *Periplaneta americana*, are innervated by dopaminergic and serotonergic nerve fibers. Serotonin stimulates the secretion of a protein-rich saliva, whereas dopamine causes the production of protein-free saliva. This suggests that dopamine acts selectively on ion-transporting cells (peripheral cells within the acini and duct cells), and serotonin acts on the protein-producing central cells of the acini. We investigated the pharmacology of the dopamine-induced secretory response of the salivary gland of *P. americana* using several dopamine receptor agonist and antagonists. The effects of dopamine could be mimicked by the non-selective dopamine receptor agonist 6,7-ADTN but not by the vertebrate D1 receptor-selective agonist SKF 38393. R(+)-Lisuride induced a secretory response but with a slower onset and a lower maximal response compared to dopamine-induced secretion. However, lisuride-stimulated glands continued secreting saliva even after lisuride-washout. This indicates that lisuride might be a partial agonist with very high affinity but low efficacy. Saliva secreted after stimulation with ADTN or lisuride had no measurable protein content arguing against serotonin receptors being activated by these drugs. Dopamine-induced secretions could be blocked by the vertebrate dopamine receptor antagonists *cis*(Z)-flupentixol, chlorpromazine and (+)-butaclamol with a similar potency.

Dynamics of biogenic amines in the brain of bee under stress conditions

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Stress treatments, CO_2 anesthesia, chilling anesthesia and vertical spin, were applied to examine whether the honeybee workers express stress responses in foraging behavior and to measure biogenic amines level changes in the stressed bee brains. A longer time interval between back and forth the feeding place was found when bees were stress-treated, suggesting that the worker foraging behavior was affected by the stress treatments. Further analysis to the stressed bee brains found that the dopamine (DA) and octopamine (OA) levels significantly decreased after chilling anesthesia but only DA was significantly decreased if the bees were anesthetized by CO_2. Similarly, the vertical spin with the velocity up to 50 and 60 rpm for 90 s, the DA and OA levels also significantly decreased. This result shows an opposite trends from the blood levels of OA and DA as reported previously, while none of these stress treatments induced changes of the serotonin (5-HT) levels neither in the blood level nor brain level, suggesting both DA and OA are the crucial compounds for stress responses in foraging behavior.

Neuroendocrine regulation of the termination of adult reproductive diapause in two species of nymphalid butterflies

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The Asian comma butterfly, *Polygonia c-aureum*, exhibits seasonal dimorphism (summer and autumn morphs) whose development is determined by photoperiod and temperature during the larval stages in a close relation to the determination of adult reproductive diapause. The physiological mechanism underlying the photoperiodic control of seasonal morph development involves a cerebral factor, named summer-morph-producing hormone (SMPH). A neuropeptide showing the SMPH-activity can be extracted with 2% NaCl and quantified by *Polygonia* pupal assay. Autumn-morph females of *P. c-aureum* and females of univoltine butterfly, *Nymphalis xanthomelas japonica*, enter adult reproductive diapause. Diapausing female adults of these two butterfly species started ovarian maturation, a termination sign of diapause, when they were treated with juvenile hormone-I (JH-I) or 2% NaCl extracts of adult brain-suboesophageal ganglion (Br-SG) complexes. Ovarian maturation was also induced by JH-I in decapitated females and isolated female abdomens of these two butterfly species. Neuroendocrine factors stimulating ovarian maturation in diapause female adults of the same species may exists in adult Br-SG complexes. Activity of the factor extracted from the adult Br-SG complexes with 2% NaCl can be assayed using female adults of the same butterfly species.

Photoperiodic regulation of the maintenance and Termination of adult reproductive diapause in the Asian comma butterfly, *Polygonia c-aureum* L.

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The Asian comma butterfly, *Polygonia c-aureum* L., exhibits seasonal polymorphism and reproductive diapause, which are controlled by photoperiod and temperature imposed during the larval (the 2nd to 5th instar) and the larval-pupal (the 2nd instar to the middle of pupal) stages, respectively (Endo et al. 1992). Larvae raised up from the egg stage under long-day conditions at 20°C develop into summer morph adults, females of which produce eggs within four days after adult emergence. Under short-day conditions at 20°C larvae develop into autumn-morph adults, females of which do not start ovarian maturation and enter reproductive diapause after adult emergence. When subjected successively to short-day conditions at 20°C, autumn-morph female adults did not start ovarian maturation more than three months after emergence. However, they were found to start ovarian maturation within 10 days and produce eggs within 15 days when they were transferred to a long-day conditions. It is concluded that adults of *P. c-aureum* count the numbers of long and short days after adult emergence, on which the maintenance and termination of their reproductive diapause may be regulated to start ovarian maturation at an adequate season.

Synaptic circuitry in the mushroom bodies of the honeybee *Apis mellifera*

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Insect mushroom bodies (MBs) are brain centers, involved in the control of complex behavior. We studied synaptic connections within the MB lip region of the honeybee *Apis mellifera* using electron microscopy combined with double labeling of the MB sensory and inhibitory input neurons. A medial antenno-cerebral tract neuron (m-ACTn) was with dextran-conjugated neurobiotin (Micro-Ruby), and GABA-ergic inhibitory neurons were labeled using immunocytochemistry. Three-dimensional reconstruction of labeled profiles showed that the m-ACTn and GABA-immunoreactive neurons (GABA-irns) had extensive contact area within the neuropil. Output synapses from the m-ACTn and GABA-irns into putative intrinsic mushroom body neurons (Kenyon cells) were revealed. GABA-irns also exhibited occasional output synapses into the m-ACTn. Our results indicate the presence of both pre- and post-synaptic inhibition within the MB calyces. The small number of inhibitory synapses on the m-ACTn may reflect a low level of ACTn inhibition within the MBs; alternatively, the level of presynaptic inhibition may vary among different subpopulations of the ACTns.

A bug rambles in the outbound trip: the navigational strategy in the *Parastrachia japonensis* (Heteroptera: Cydnidae)

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A female of subsocial bug, *Parastrachia japonensis* (Heteroptera: Cydnidae) transports drupes of a host tree to a nymph-containing burrow on the ground by the host tree. Because its burrow usually located up to 5m away from the host tree, the bug should take the round trip between its burrow and the area where the drupes could be obtained. We compared the outbound trip with the homing one and there was much difference. In the first outbound trip, the bug oriented randomly and searched tortuously, however, it failed many times to obtain a drupe. When the bug had encountered one, it always took the shortest route back to the burrow. After the successful trip, the bug oriented toward the position where it had obtained the drupe in the previous navigational trip using the path integration. However, its outbound trip was slower and unsteadier than the homing one. The burrow is a limited goal for the homing bug, whereas the drupes as goals for the foraging bug distribute over the ground with several patches. The behavioural differences of the outbound trip from the homing one are assumed that the bug changes the navigational strategy adjusting the spatial configurations of their goals.

Partial purification of *Bombyx* cerebral peptide showing activity of summer-morph-producing hormone (SMPH) in the Asian comma butterfly, *Polygonia c-aureum* L.

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Asian comma butterfly, *Polygonia c-aureum*, exhibits seasonal (summer and autumn) morphs. Physiological mechanism underlying the photoperiodic control of seasonal morph development involves a cerebral factor, named summer-morph-producing hormone (SMPH). The cerebral factor is a peptide which can be extracted from brains of *Polygonia* long-day pupae with 2% NaCl and purified partially by a gel-filtration and a reverse-phased HPLC on *Polygonia* pupal assay for the SMPH. Besides, a cerebral peptide showing SMPH-activity in *P. c-aureum* was found to exist in brain-suboesophageal ganglion (Br-SG) complexes of the silkmoth, *Bombyx mori.* The peptide could be extracted and purified partially by the same manner as above (Tanaka et al., 1997). Here, we tried to extract the SMPH-active peptide from Br-SG complexes of *B. mori* adults and were purified partially by a gel-filtration with Superose-12 column and 6 steps of reverse-phased HPLC with C-8, C-4 and C-18 ODS-column. We succeeded to obtain highly purified SMPH-active peptide as a single peak fraction in absorbance at 220 nm.

Immunoreactivity of glutamic acid decarboxylase (GAD) isoforms in the central nervous system of the barn spider, *Araneus cavaticus*

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The γ-aminobutyric acid (GABA) has long been considered an inhibitory neurotransmitter in the central nervous system (CNS) of both vertebrates and arthropods. The glutamic acid decarboxylase (GAD) catalyzes the conversion of L-glutamate to GABA. As the GAD has a restricted tissue distribution, it is highly expressed in the cytoplasm of GABAergic neurons in the CNS. However it is also present in other non-neuronal tissues such as testis, oviduct and ovary. Recently, there were reports that a GABA-like immunocytochemical reactivity and a ninhyin-positive GABA derivative, GABamide, exists in the visual ganglia and in the water-soluble fraction of the spider web respectively. So, this experiment initiated to identify exact distribution of the GAD isoform immunoreactivity in the CNS of the spider to reveal the ecophysiological significance of GABA for spider's behavior.

Cloning of biogenic amine receptor fragments from *Periplaneta* and *Schistocerca*

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Biogenic amines regulate and modulate many physiological and behavioral processes in insects. The identification of biogenic amine receptors is decisive for an understanding of the cellular pathways regulated by the amines. Biogenic amine receptor genes encode membrane proteins that primarily belong to the superfamily of G protein-coupled receptors (GPCRs). GPCRs share the common motif of seven transmembrane domains (TM). We endeavoured to isolate members of this gene family from two hemimetabolous insects, the American cockroach, *Periplaneta americana*, and the desert locust, *Schistocerca gregaria*. Both species are important model organisms for neurobiological and physiological research. Degenerate primers were designed corresponding to highly conserved sequences in TM 6 and 7 of already cloned insect receptors. Using a *P. americana* brain cDNA library and *S. gregaria* genomic DNA as templates, four and three different PCR fragments were amplified, respectively. BLAST similarity searches to Genbank showed that the cloned fragments are derived from different biogenic amine receptor genes of both species.

Antagonists of $5HT_3$-like receptors of invertebrates as pesticides

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Serotonin (5HT) modulates pharyngeal pumping in nematodes. This paper concerns novel pesticides acting on $5HT_3$-like receptors in both nematodes and insects. Two selective and structurally unrelated antagonists of the vertebrate $5\text{-}HT_3$ receptor, MDL-72222 (tropanyl dichlorobenzoate) and ondansetron, were tested for their effects on feeding and survival of nematodes and insects. When applied to the nematode *Caenorhabditis elegans*, MDL-72222 stops pharyngeal pumping. MDL-72222 also causes dose-and time-dependent mortality of free-living and parasitic nematodes. The IC_{50} of MDL-72222 for blocking pharyngeal pumping of the nematode *C. elegans* was 10 M. The LD_{50} of MDL-72222 for the murine hookworm *Heligmosomoides polygyrus* was 19 M (3 days) and 5 M (5 days). Statistically significant mortality was also observed against the peach-potato aphid, *Myzus persicae*. MDL-72222 had little or no effect on growth rate of the caterpillar, *Helicoverpa armigera*. Ondansetron had qualitatively similar effects to MDL-72222 but at higher concentrations. Chemical analogues of MDL-72222 were synthesised and tested against *C. elegans* and *H. polygyrus*. One showed slightly higher potency against nematodes than MDL72222. We also report development of a 96-well-plate based assay for detecting compounds which affect pharyngeal pumping of nematodes. Patents pending.

Does the neuroanatomy of visual neurons in hoverfly match visual ecology?

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Wide-field (tangential) neurons of the fly lobula plate collate information about motion. One class of such neurons is the 'horizontal system' or HS cells. These respond with graded, direction-selective responses to wide-field motion associated with yaw during flight. In several families of Cyclorraphan flies (Calliphoridae, Muscidae and Drosophilidae) there are believed to be 3 HS cells in each optic lobe. These cells divide the visual world into 'north' (HSN), 'equatorial' (HSE) and 'south' (HSS) regions. Previous work suggests that the number and morphology of HS neurons is variable amongst other species of dipteran flies in a manner that consistent with visual ecology. However, there is little supporting evidence for functional differences amongst fly species from electrophysiology. The hoverfly, *Eristalis*, is emerging as a physiological model for the study of such motion detecting neuron, and exhibits spectacular aerial performance compared with other Cyclorraphan species. We have commenced electrophysiological recordings with detailed anatomical investigation of these neurons using intracellular injection with fluorescent probes (Lucifer yellow, Alexa dyes) and confocal microscopy. Our initial data suggest that *Eristalis* has additional HS neurons, dividing the world into smaller segments than in the other Cyclorraphan species studied to date.

Characterization of small motion detection neurons of *Eristalis tenax*

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The hoverfly *Eristalis tenax* exhibits typical syrphid behaviour, with stationary hovering followed by rapid pursuit of conspecifics. Male and (possibly) female flies depend on detection of small rapidly moving targets to find mates and defend their territory. Neurons specialized for detecting small targets against cluttered backgrounds can be found in the lobula complex (3rd optic ganglion) of the fly brain. We are characterizing the morphology and physiology of a range of lobula small target motion detecting (STMD) neurons using dye-fills and electrophysiological techniques. We define STMDs as neurons that give a powerful response to a small object (1-2 degrees square) moving across a bright or cluttered background, but weaker responses to larger size targets. We find a number of classes of STMDs with unique properties in both sexes. These include neurons with small excitatory receptive fields surrounded by distinctly shaped inhibitory areas, and wide-field STMDs that may be used for mate detection. STMDs generally give spiking responses, and have variable degrees of spontaneous spike activity, including neurons that are 'silent' until stimulated by a target. A prominent feature of some STMDs is the robustness of their responses in the presence of background motion.

Comparisons of the image-step response in flies & humans

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Neurons specialized for detecting motion patterns are located in the fly lobula complex (3rd optic ganglion). HS (horizontal system) cells give graded direction-selective responses to the stereotypical wide-field motion generated during flight. It has long been known that identical cellular responses can be generated by apparent motion, e.g. by a stationary image that jumps in the preferred direction. HS cells give a strong, sustained response over a few milliseconds to stimuli of this kind, where an adapting image appears and then steps a few degrees in either direction (anti-preferred direction gives an inhibitory response). The amplitude of the response depends on factors such as the contrast and duration of the adapting stimulus. Human subjects do not seem to perceive the image-step stimulus as continuous motion, despite the fact that many other visual motion illusions have been shown to be similar in fly wide-field cells and human perception. By comparing fly electrophysiological data with human psychophysics we are analysing the cause of these different responses.

Pattern noise in insect motion detector responses to natural scenes

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We are using the Hoverfly *Eristalis* as a physiological model for studying visual motion analysis in natural scenes. Natural images have predictable statistics, and biological systems may be optimized to take advantage of this predictability. In insect vision, outputs of local motion detectors are collated by a system of tangential neurons tuned to very specific types of complex motion. We utilized intracellular recordings from tangential (HS) neurons stimulated by natural panoramic images in an open-loop yaw simulator. Differences in the local structure of natural images result in variations in the correlated response of the local detectors. We investigated this 'pattern noise' through experiments, in which we present images at various initial positions and versions of the same images modified to disrupt vertical contours. By aligning responses in time across several initial positions, pattern noise is largely removed from the mean, but contributes to standard deviation. We propose a computational model with additional nonlinearity (saturation, motion adaptation) based on this principle and taking account of the receptive field shape of the tangential neurons. Such a model can provide a robust indication of the angular velocity of optical flow, with widespread applications in artificial vision systems.

Radar observations reveal the efficacy of path integration mechanisms in flying bees

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We have used harmonic radar to record the complete flight paths of individual foraging honeybees that had been trained to a feeder 200 m from their hive. When the feeder was removed, hive-departing bees maintained accurate flights (often in strong gusty cross-winds) to where the feeder had been, and then searched over a progressively larger area centred on this position, before eventually returning to the hive. In spite of the extensive searching phase, the final approach to the hive was often impressively accurate, demonstrating that the bees' combination of path integration and wind compensation was remarkably robust. Over the relatively short distances and open landscape of the present experiment, changing the position of a prominent landmark adjacent to the hive-feeder axis produced no discernible effect on either the outward or return 'vector' flights. This indicated that the bees were relying entirely on path integration, and flying in an 'automatic pilot mode' seen in bees that have been artificially displaced.

Turn alternation in the pill bug on a servosphere

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In a maze consisting of a series of T-branched trenches meeting at right angles, a pill bug, *Armadillidium vulgare*, generally chooses branches in an alternative way: once it has turned right at a branching point, it will turn left at the next. The *turn alternation* response, which has been considered to be due to short-term memory regarding the turn-direction or compensatory leg-movement after the previous turn, was re-investigated in a T-maze as well as on a *servosphere* locomotion compensator. Because of the intense thigmotaxis, the animals were contacting and following either the right or left wall of the trench to a branching zone, where they move away from the wall and strayed into the zone. Since most of the animals made a slight turn towards the contacting side at leaving the wall, they hit the facing wall with the contralateral appendages then started wall-following with the other side of the body. This behavioral context, being reproduced on a servosphere with control of tactile cues, cause a pill bug to make alternating turns in a T-maze.

Suboesophageal neck motoneurons in the honeybee, *Apis mellifera*

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We are interested in how the visual input during flight and other locomotion is translated into corrective movements of the head, body and abdomen in the honeybee. We studied the morphology of neck motoneurons (NMs) that run in nerve IK1 and innervate neck muscles 44 and 51. Muscle 44 is a direct neck muscle, which serves to pull the head downwards, muscle 51 is an indirect muscle of undetermined function. Recordings from IK1 have shown that these NMs are motion-sensitive. With dye-application into muscle 44 we labeled several NMs in the suboesophageal ganglion, where they innervate the dorsal neuropil, an area that is also innervated by motion-sensitive descending interneurons. A group of NMs that look very similar was stained via dye-application into muscle 51: two NMs with a median cell body and two NMs with a lateral cell body. To find out, whether muscles 44 and 51 are innervated by the same or different NMs, we investigated the number of neurons that run in nerve IK1. In cross sections of IK1 we found eight axon profiles larger than 10 µm. The data suggest that muscles 44 and 51 are innervated by different, but similar NMs.

Processing of mosquito signaling peptides by ACE: a functional genomics approach.

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Neuropeptides are important regulators of many aspects of physiology and development. Hydrolysis by extracellular peptidases can result in activation; alteration of biological activity; or inactivation. Mammalian angiotensin-converting enzyme (ACE) is one of the best known metallo-enzyme peptidases. ACE homologues have been in found in all insect species studied so far. Two *Drosophila melanogaster* ACE genes (*Ance* and *Acer*) are known to code for proteins with peptidase activity. The expression pattern and mutant phenotypes of these genes suggest important roles for these enzymes at different stages of development. ACE activity has been shown to rise after a blood meal in the mosquito *Anopheles stephensi* where it may play a role in control of host blood pressure, in regulating peptide activity after a blood meal or as a means of compromising host defense mechanisms. We have identified 9 ACE-like genes in the mosquito *Anopheles gambiae*. We aim to identify full length cDNA clones for these AnoACE genes and express the AnoACE proteins in the yeast *Pichia pastoris*, in order to investigate functional conservation of this group of enzymes in insects, to understand the physiological role of *A. gambiae* ACE, to and to identify selective inhibitors of ACE with potential for vector control.

Comparative genomics of *Drosophila* odorant receptors

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The odorant receptors (Ors) of *Drosophila melanogaster* are a highly divergent family of 62 seven trans-membrane domain proteins, presumed to be G-protein coupled receptors, that exhibit no primary sequence similarity to odorant receptors from nematodes, vertebrates, or to any other family of GPCRs. In several families of GPCRs conserved regions in intracellular loops 2 and 3 and the C terminus interact with G proteins. However in the diverse *D. melanogaster Or* family (average level of identity ~20%), such conserved regions are difficult to identify. We are therefore using a comparative genomics approach to identify domains in *Drosophila* Ors that may interact with downstream signalling components. We have isolated orthologues of nine different *Or* genes from other members of the *melanogaster* subfamily. Individual sequence identity among the species ranges from 60-98% at the amino acid level. The number of exons and introns and the position of introns are conserved in all species, however intron sizes vary. This data has helped us identify potential functional regions, which will be described. We have also used this data to undertake analysis of the evolutionary dynamics and patterns of selective pressure on these genes, and these results will be presented.

Are innate colour preferences adaptive for young forager bees?

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Bees show high flexibility in colour learning. However, testing of colour-deprived bees revealed an inherited colour memory. Unexperienced foragers exhibit colour preferences toward targets appearing to humans blue or violet. They are suppressed by colour experience acquired during nectar foraging but may interact with decision making in a novel context. We focused on the question whether such colour preferences might be adaptive for young bee foragers by studying the distribution of bee-subjective colour cues and nectar properties in natural grassland habitats. We hypothesised blue-violet flowers to be either highly rewarding or most abundant. The flower colours of 10-15 bee-pollinated and key plants were measured in each of several natural patches. Bee-subjective spectral parameters were determined using the RNL model of honeybee colour vision. Simultaneously we recorded insect visitation, nectar properties and floral abundances. Flower colours were widely distributed in the perceptual colour space representing different colour hues to bees. High abundance and reward were characterised for blue-violet flowers but also for several other colours. The flowers with highest colour contrast, possibly facilitating the identification as floral object, were not exclusively blue-violet. We suggest that innate colour preferences found in higher bees might be related to physiological or phylogenetical constraints.

Photoperiodic and thermal regulation of pupal beige/black polyphenism in the small copper butterfly, *lycaena phlaeas* Daimio Seitz

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The small copper butterfly, *Lycaena phlaeas* Daimio Seitz, has pupal beige/black polymorphism which was controlled by photoperiod and temperature imposed during the larval stages in an apparent association with the development of adult (spring and summer) morphs. Under long-day conditions at 23°C larvae developed into beige-type pupae and into red-winged adults, whereas under short-day conditions larvae developed into black-type pupae and into brown-winged adults. When young pharate pupae obtained under long-day conditions at 24°C were subjected to 4°C for 5 days, they develop into TB-pupae, head-thoraxes and abdomens of which were judged as to be black and intermediate types, respectively. Adults developed from the TB-pupae were found to have reddish wings as compared to the unchilled (control) ones. The results indicate that the physiological mechanism underlying the photoperiodic control of seasonal morph development may play a significant role in the determination of pupal beige/black polyphenism in *L. phlaeas daimio*. Furthermore, melanization of the pupal cuticle may occur in a close relation to the determination of the color of adult wings.

Spectral sensitivity & color preference of the oriental fruit fly, *Bactrocera dorsalis* (Diptera: Tephritide)

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Several species of fruit flies (Tephritidae) are the destructive agricultural pests widespread around the world, and the oriental fruit fly, *Bactrocera dorsalis*, is the critical one in Pacific Rim. Numerous "yellow traps" are applied to control this pest; however the crucial attractive cues have still not been fully investigated. In this study, the spectral sensitivities of the photoreceptors in the compound eyes of *B. dorsalis* were measured intracellularly. Five spectral types of photoreceptors with the peak sensitive wavelength at 370, 380, 490, 510 nm, and one with dual peak at 350 and 490 nm respectively were recorded, indicating the visual spectrum of the fly mainly distributes over the UV (300 ~ 400 nm) and blue-green (400 ~ 550 nm) ranges. In addition, the color preference of the fly was tested by the attractive effects of color papers with different reflectance spectra. It showed the color papers with reflectance spectra rising from 490 nm are more attractive than those rising from 560 nm, indicating that the visual stimuli with spectrum covering the range of 490 ~ 560 nm has a positive effect to attract the oriental fruit fly.

Neuroendocrine regulation of the orange coloration of diapause pupae in the swallowtail butterfly, *Papilio xuthus* L. (Lepidoptera: Papilionidae)

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Diapause pupae of *Papilio xuthus* show color polymorphism, represented by diapause-green, orange, and brownish orange types that are each associated with specific pupation sites. We investigated the role of the site of pupation on the induction of the development of orange types (or brownish-orange types), and the neuroendocrine mechanism underlying the control of color polymorphism in short-day pupae. All short-day larvae of the wandering stage developed into orange or brownish-orange type pupae when they were placed in rough-surfaced containers after gut-purge. Utilizing a pharate pupal ligation between the thorax and abdomen, the neuroendocrine mechanism underlying the control of color polymorphism was shown to involve a head-thorax factor (Orange-Pupa-Inducing Factor: OPIF) that induced orange types in short-day pupae. OPIF was bioassayed using the ligatured abdomens of short-day pharate pupae and extracted with a 2% NaCl solution from 5th-instar larval ganglia complexes following the mesothoracic complex ($TG_{2,3}$-AG_{1-7}), but it could not be extracted with either an acetone or 80% ethanol solution. OPIF may not exist in the brains of day 0 pupae or in brain-subesophageal ganglion and prothoracic ganglion complexes of 5th-instar larvae. The action of OPIF on short-day pharate pupae is considered dose-dependent.

Patterns of chromatic information processing in the lobula of honeybee

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The honeybee, *Apis mellifera* L., is one of the few living creatures that have their colour vision proven through behavioural tests. Previous studies on a honeybee's colour vision emphasized the relationship between the spectral sensitivities of photoreceptors and colour discrimination behaviour, and the knowledge regarding neural mechanisms of colour vision is rather limited. We surveyed the patterns of chromatic information processing of visual neurons in the lobula of the honeybee, using intracellular recording and three light emitting diodes, whose emitting spectra approximately match the spectral sensitivity peaks of the honeybee, as the light stimuli source. The recorded visual neurons can be divided into a non-colour opponent cells group and a colour opponent cells group. In non-colour opponent cells group, in which six response patterns of broad-band neurons and four response patterns of narrow-band neurons were recorded. In colour opponent cells group, the principal neural mechanism of colour vision, eight response patterns of colour opponent cells were recorded. Thus, together with previous studies, the visual system of the honeybee might possess 26 types of colour-coding neurons, including 12 types of colour opponent neurons. Besides, some recorded neurons with tonic post-stimulus responses were observed, suggesting spectro-temporal opponency may be part of the colour-coding mechanisms.

Colourful orb-weaving spiders, *Nephila pilipes*, through a bee's eyes

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Many orb-weaving spiders in the tropics forage in open sites during the day and some of them have both bright and dark colorations. The conspicuous UV reflective colour markings of these spiders have been reported to be attractive to visually oriented prey and thus could increase the spiders' foraging success. Using a combination of field and laboratory studies, we examine whether or not the body coloration of orb-weaving spiders exhibit optical properties that are attractive to insect prey from the viewpoints of insect visual physiology. We compared the prey interception rates and colour contrasts of the typical and melanic morph of giant wood spider *Nephila pilipes*. Results of the field study showed that the typical morph *N. pilipes* caught significantly more insects than the melanics. Colour contrasts calculated from spectral reflectances of the background and body surface of spiders showed that the brightly-coloured body parts of typical morph exhibited rather high values, but those of the dark body parts were below the discrimination threshold. The differential colour contrasts of body parts generated a visual signal unlike that of a spider but certain form of food resources. On the other hand, the melanic morphs did not have bright coloration and the colour contrasts of every parts of body were significantly higher than the threshold, making the contour of spiders quite clear to bees.

Fine structural analysis of the silk apparatus in the spider, *Paracoelotes spinitarsis* (Araneae: Amaurobiidae)

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The silk of spider is used to form various structures such as aglines, retreats, webs, snares, and cocoons. Since the interactions between spider and environment are silk-mediated, the silk apparatus and its silk glands deserve treatment from morphological and ecological point of view. The silk apparatus of this spider, *Paracoelotes spinitarsis* is composed of internal silk glands and external spinnerets. Among the three pairs of spinnerets, the posterior pairs are highly elongated along the body axis. In order to understand the properties and the evolutionary origins of these silk constructs, we analysed here the morphological characteristics of the silk spinning apparatus and its ecological significance in this spider. Through the fine structural analysis using light and electron microscopes, the silk glands of the adult female spider were composed of four types of silk glands (ampullates, tubuliforms, aciniforms and pyriforms) which connected with typical spinning tubes of each spinneret. It has been also revealed that the tubuliform glands were only observed in female spiders, however the flagelliform and aggregate glands which had the function of adhesive thread production in orb-web spiders were not observed at both sexes of this spider.

Corazonin receptor signaling in ecdysis initiation.

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Corazonin is a highly conserved neuropeptide hormone in widely divergent insects, yet is associated with no common function. Following discovery of the corazonin receptor in *Drosophila*, we identified its ortholog in *Manduca sexta*. Corazonin receptor cDNA in *Manduca* encodes a protein of 436 amino acids (MasCRZR) with seven putative transmembrane domains and shares common ancestry with its *Drosophila* counterpart. MasCRZR exhibits high sensitivity and selectivity for corazonin when expressed in *Xenopus* oocytes (EC_{50} ~200 pM) or Chinese hamster ovary (CHO) cells (EC_{50} ~75 pM). Northern blot analysis locates its expression in peripheral endocrine Inka cells and in the CNS. Injection of corazonin into pharate larvae elicits release of pre-ecdysis and ecdysis triggering hormones (PETH and ETH) from Inka cells, followed by precocious pre-ecdysis and ecdysis behaviors. Corazonin (25-100 pM) also induces PETH and ETH secretion from isolated Inka cells *in vitro* and circulates in the hemolymph (20-80 pM) just prior to natural pre-ecdysis onset. Early and late phases of peptide release from Inka cells, controlled by corazonin and eclosion hormone, respectively, appear to regulate sequential steps in the ecdysis behavioral sequence.

PTTH signal transduction in the prothoracic gland: a tangled web with one purpose?

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Molting and metamorphosis in insects is controlled by several hormones, with 20-hydroxyecdyone (molting hormone) playing an overarching role. Prothoracicotropic hormone (PTTH), the best-understood regulator of ecdysteroidogenesis, stimulates ecdysteroid synthesis by the prothoracic glands. Work in the 1980s and 1990s revealed that PTTH activated a signal transduction cascade that included Ca^{2+}- influx, cAMP generation, phosphorylation of the ribosomal protein S6 and protein translation. In this talk, I will review more recent work, from our group and others, indicating that the PTTH signal transduction cascade is surprisingly complex. We now know that additional kinases are activated, including an extracellular signal-regulated kinase (ERK), a protein kinase C, and one or more unidentified tyrosine kinases. Ongoing studies also suggest PTTH-stimulated Ca^{2+}- influx is complexly regulated, with increases in cytoplasmic free Ca^{2+} coming from at least three sources, and that phospholipase C activation is among the earliest intracellular events triggered by PTTH. The composite data suggest that (1) PTTH regulates more processes in the prothoracic gland than ecdysteroid synthesis; (2) the study of phosphorylated proteins offers an avenue to identify these additional functions of PTTH; and (3) the control of ecdysteroid synthesis is subject to a Pandora's box of developmentally varying checks and balances.

Dielectric waveguide antenna theory helps explain insect detection of enantiomers

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Many decades ago, a debate arose as to how insects detected semiochemicals. One group maintained that detection was accomplished by electromagnetic energy emanating from the semiochemical, while the other group maintained that the molecule fit into a receptor in order for it to be detected. The latter group's theory is termed the "lock and key model". It is the predominant theory today. The former group's theory was termed the "vibrational theory of odor" by Robert Wright in the 1950's. In the 1960's, Philip Callahan offered a mechanism for this detection whereby the insect sensillae act as dielectric antennae. Behavioral and electrophysiological tests involving the detection of enantiomers could not be easily explained by either theory. Some insects could differentiate between enantiomers while others could not. A more thorough knowledge of antenna theory reveals that enantiomer detection can be explained by the dielectric antenna model. Additionally, replicas of insect sensillae can be artificially reproduced by conventional lasers to make efficient infrared detectors.

"Insects as Flying Machines"

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Insects are small flying machines with an apparent disregard of good aerodynamic practice. Instead of producing lift with their wings, some butterflies rely on drag for slow flight. Tiny insects were predicted to 'swim' through the air using drag, but they rely more on lift. Hovering dragonflies and hoverflies flap their wings in a plane inclined to the horizontal. They use dynamic stall on the downstroke to support their weight, and their lift-to-drag ratio is necessarily less than 2. Most hovering insects flap their wings in a horizontal plane, using lift on both the downstroke and upstroke to support their weight. A leading-edge vortex created by dynamic stall enhances lift production. Because of flow separation at the leading edge, the resultant aerodynamic force is approximately normal to the wing surface, and the lift-to-drag ratio is again typically less than 2. The lift-to-drag ratios for all high-force mechanisms employed by insects are appallingly low by conventional standards, and would entail a high power consumption for fixed and rotary wing designs. For small man-made flying machines to exploit these mechanisms, a flapping motion is essential to ensure that weight is supported by the resultant aerodynamic force and not just the lift.

Controlling flapping flight: lessons from locusts, dragonflies and *Aedes* mosquitoes.

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"To a flying insect, unsteadiness is everything. With wings flapping tens or hundreds of times every second, the airflow changes continually, but despite – or perhaps because of – this complexity, insects are able to control manoeuvres with greater finesse than even the highest performance fighter aircraft. By adopting and adapting techniques used by engineers to analyse aircraft stability and control, it is possible to predict an insect's response characteristics to accidental disturbances and deliberate control inputs. By analysing the insect's natural modes of motion, we can then make quantitative predictions about the operating bandwidth of the nervous system and even the preferred directions and temporal resolution of its sensors. Besides yielding integrative physiological insight, analysing the insect as a miniature flying machine is instructive for the engineer seeking to mimic aspects of insect flight. In this talk I will illustrate some of the lessons we can learn from behaviours as diverse as the cruising flight of locusts, the take-off manoeuvres of dragonflies, and the mating flights of *Aedes* mosquitoes. Finally, I will show how an understanding of flight control can even contribute to pest control, by helping us select the most successful male mosquitoes for use with the sterile insect technique.
"

What drives adaptive gain control in insect motion detection?

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"The phenomenon of motion adaptation is widespread among animals, having been demonstrated in species as diverse as humans, wallabies and flies. Motion adaptation appears to have evolved to maximize efficiency of visual coding in neurons with limited signaling range. Recent work highlights controversy over the site and mechanism of adaptation. Hypotheses range from simple gain control in 'early' vision (contrast normalization), to adaptive feedback that alters the delay mechanisms within local motion detectors. We have shown that adaptation does not alter the velocity tuning of motion detectors, but instead induces large reductions in contrast gain. Using moving natural scenes and a probe-adapt-probe protocol, we show here that gain reduction is driven by a mechanism with broad velocity tuning. While the tuning of adaptation resembles that of motion detectors themselves (favoring a feedback hypothesis), its broadness suggests extremely high gain in the feedback pathway and permits powerful adaptation to stimuli that drive motion detectors weakly. This mechanism may optimize coding of image velocity independent of the contrast variations between different natural scenes. Our work suggests easily implemented elaborations to existing motion detector models that could impart similar robustness to artificial motion detectors based on the same principle.
"

"Frictional properties of contacting surfaces in the hemelytra-hindwing locking mechanism in the bug *Coreus marginatus* (Heteroptera, Coreidae)"

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The structure and function of the hemelytra-to-hindwing locking mechanism of the bug *Coreus marginatus* were analysed. The system consists of a cuticular protrusion in the ventral side of the hemelytra, which locks the subcostal border of the hindwing in flight. The speed and distance slid by both surfaces against one another during flight were assessed using a combination of high-speed video recordings and a 2D geometrical model. The friction coefficient between sliding surfaces was assessed using a micromanipulator, coupled with force transducers. This was done under three experimental conditions: freshly dissected, air dried and rehydrated ethanol preserved samples. The results showed a high speed of sliding, approximately 0.18 $m \cdot s^{-1}$, with a relatively low friction coefficient (0.2 µ). The surface morphology of this mechanism, namely outgrowths of one part having rounded edges, and completely flat surface on the counterpart, effectively aids in the reduction of friction at the microscopical level. The structure is effective even dry, and after being preserved in ethanol, suggesting that no cuticle secreted lubrication substance is responsible for its effectiveness. The ultrastructure presumably confers mechanical stability to the system under the high load it is subjected to in flight.

Potential Northern Distribution of *Anoplophora glabripennis* (Motschulsky) in North America

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The Asian long-horned beetle (alb), *Anoplophora glabripennis*, is a cerambycid endemic to China and the Korean peninsula that was recently introduced into North America. As a result, it now infests the urban forests of Long Island, Chicago, New Jersey and Toronto. Most North American insects from this family are usually consider beneficial because they hasten the breakdown of dead trees. However, members of the genus *Anoplophora* infest healthy hardwood trees. The preferred tree species in China are *Salix* and *Populus* whereas in North America *Acer* species are most commonly attacked. The supercooling point of insects has been used to predict the potential northern distribution of insects susceptible to freezing. These measurements have given a reasonably accurate prediction of the northern distribution of the gypsy moth and the smaller European elm bark beetle in North America. While there is no assurance this is an accurate assumption because of insect physiological and behaviorally adaptations, it does provides a useful tool for estimating the potential northern distribution of newly introduced species unless that species is freeze-tolerant. In our study, we investigated the supercooling point of alb larvae, its potential impact on distribution in North America and how this compares to a native cerambycid.

"Antioxidant and detoxifying enzyme activities in two strains of *Oncopeltus fasciatus* (Heteroptera: Lygaeidae) exposed to cadmium: I. Acute exposure."

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"Catalase (CAT), general esterase (EST), glutathione reductase (GR), glutathione S-transferase (GST), peroxidase activity of glutathione S-transferase (GSTpx), and superoxide dismutase activity (SOD) were determined in adult *O. fasciatus* exposed to Cd in drinking water for five days. Two different strains were studied: OS, a laboratory-reared strain, and OCdR, a strain subjected to Cd selection. In order to distinguish between genetic and physiological effects, two generations of OCdR were used in the experiments. OCdR F_{49} was born from 50 mg Cd/ml selected parents, but was not exposed to Cd itself, while OCdR F_{50} was exposed to 50 mg Cd/ml from egg eclosion to the moment of the experiment. Median lethal mortalities (LC_{50}) of adults after 5 day-exposure were 360 mg Cd/liter for OS, 412 mg Cd/l for OCdR F_{49}, and 455 mg Cd/litre for OCdR F_{50}. Survivors from toxicity bioassays exposed to 0, 100 and 200 mg Cd/litre were used for enzyme assays. CAT activity was significantly enhanced in Cd-treated insects of all strains, when compared to control insects of the same group. EST, GR and GST activities where only significantly enhanced in insects exposed to the highest Cd concentration, while GSTpx and SOD activities were not affected by Cd treatment. On the other hand, no differences in basal levels of enzyme activities were observed among strains.
"

The visual ecology of hawkmoths

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The diurnal *Macroglossum stellatarum* and the nocturnal *Deilephila elpenor* are active in amatically different light intensities but both have colour vision. *D. elpenor* can discriminate colours in light intensities a million times weaker than the light intensities normal for *M. stellatarum*. Both species have colour constancy, which allows them to discriminate colours irrespectively of the spectrum of the illumination. This ability is explained by chromatic receptor adaptation according to the von Kries coefficient law. *M. stellatarum* learns the size, patterns and brightness of flowers. Both species learn the spatial location of rewarding stimuli, when they are difficult to distinguish by colour alone. Nocturnal sphingids mostly use odour to localize nectar flowers. However, in the day-active *M. stellatarum*, vision has become the dominating modality. When faced with a colour and an odour stimulus, naive *M. stellatarum* select the colour. The behaviour of flower-naive *D. elpenor* is the opposite. We are currently investigating the role of multi-modal interaction in learning experiments with both species.

Technical perfection in the sensory periphery: spider mechanoreception

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Despite the small size of their brain arthropods show complex and highly refined behavior. One of the foundations making this possible is the refinement and filter action of their sensory periphery. Spider mechanoreceptors exemplify this nicely. Being interfaces between environment and behavior they not only reflect details of the physical properties of the adequate stimulus but also characteristics of species-specific behavior. This will be illustrated taking two different types of sensory hairs as examples. (i) *Trichobothria* are *medium flow sensors* of outstanding sensitivity which is largely due to an intriguingly well "designed" interaction between the moving air and the hair. Using fluid mechanics this interaction can be modelled quantitatively and the hair´s mechanical and nervous responses related to prey catching behavior. (ii) *Tactile hairs* follow mechanical principles different from those of a medium flow sensor as seen from their morphology and mechanical properties and revealed by finite element analysis and micromechanical measurements. Bringing together neurobiology and engineering not only deepens our understanding of sensory systems but also helps in developing new, biology-inspired technical sensors.

Molecular and pharmacological characterization of *Drosophila* octopamine receptors

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In invertebrates, the biogenic amine octopamine regulates physiological functions including locomotion, neuronal development, circadian rhythm, as well as learning and memory. Pharmacological investigations indicate that octopamine mediates these effects by activation of several G protein-coupled receptor types.
We have cloned two genes from *Drosophila melanogaster* that encode three functional octopamine receptors. The first gene (Dm*oa*1) codes for two polypeptides which are generated by alternative splicing. The proteins share identical N-terminal halves but differ in the C-terminal half. When stimulated with nanomolar concentrations of octopamine, both splice variants induce Ca^{2+} release from internal stores. At high concentrations of octopamine (${}^{3}10^{-6}$ M) the receptors also cause cAMP production. The second gene (Dm*oa*2) encodes a single polypeptide that specifically causes a rise of intracellular cAMP. The EC_{50} value is ~5×10^{-8} M octopamine. Tyramine activates all three receptors but at ^{3}100 fold higher concentrations. The biogenic amines dopamine and serotonin are non-effective at all. Developmental expression of Dm*oa* genes was assessed by RT-PCR. Overlapping but not identical expression patterns were observed for the individual transcripts. Since native octopamine receptors induce either cAMP production or Ca^{2+} release, the two genes may encode the complete set of octopamine receptors that are expressed in *Drosophila*.

The imaging properties of the dragonfly median ocellus

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Current literature describes the ocelli of pterygote insects as average illumination detectors, which serve to provide attitude information for the maintenance of flight equilibrium. This view has arisen because of several prominent features of the ocelli, including; fields of view centred on the horizon, under-focussed optics and a high rate of convergence from photoreceptors to second-order neurons which output to regions of the nervous system involved in flight control. However, recent research has indicated that dragonfly ocelli possess unusual structural and optical properties that are inconsistent with this view of ocellar function. In particular, from hanging-op preparations of the dragonfly median ocellar lens it has been determined that the image formed by this lens lies close to the underlying retinal layer, that the resolution of the image is dependent upon the orientation of an object and that the image is split laterally into left and right halves. Preliminary investigations also indicate the presence of a gradient of refractive index in the median ocellar lens. The optical properties of this lens and their relevance to dragonfly behaviour will be discussed.

Stop and go with NO: nitric oxide and cyclic nucleotides regulate neuronal motility in an insect embryo

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The dynamic regulation of nitric oxide synthase (NOS) activity and cyclic GMP levels suggests a functional role in the development of nervous systems. Here, we report evidence for a key role of the NO/cGMP signalling cascade on migration of postmitotic neurons in the enteric nervous system of the embryonic grasshopper. During development, a population of enteric neurons migrates on the surface of the midgut. These midgut (MG) neurons exhibit nitric oxide-induced cGMP-immunoreactivity coinciding with the migratory phase. Using a histochemical marker for NOS, we identified potential sources of NO in subsets of the midgut cells below the migrating MG neurons. Inhibition of endogenous NOS, sGC, and protein kinase G (PKG) activity in whole embryo culture results in a significant retardation of MG neuron migration. This inhibition can be rescued by supplementing with protoporphyrin IX free acid an activator of sGC, and membrane-permeant cGMP, indicating that NO/cGMP signalling is crucial for MG neuron migration. Conversely, the stimulation of the cAMP/protein kinase A signalling cascade results in an inhibition of cell migration. Activation of either the cGMP or the cAMP cascade influences the cellular distribution of F-actin in neuronal somata in a complementary fashion. The cytochemical stainings and experimental manipulations of cyclic nucleotide levels provide clear evidence that NO/cGMP/PKG signalling is permissive for MG neuron migration whereas the cAMP/PKA cascade may be a negative regulator. Since NO/cGMP signaling is also involved in axonal extension of pioneer neurons, these investigations reveal an accessible insect model in which the role of the NO/cGMP/PKG signalling cascade on neuronal cell motility can be analyzed in a natural setting.

Excitation-secretion coupling in the salivary glands of *Periplaneta americana*

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The acinar salivary glands of the cockroach, *Periplaneta americana*, are innervated by dopaminergic and serotonergic nerve fibers. Serotonin stimulates the secretion of protein-rich saliva, whereas dopamine causes the production of protein-free saliva. This suggests that dopamine acts selectively on ion-transporting peripheral cells within the acini and the duct cells, and serotonin acts on the protein-producing central cells of the acini. We investigated the pharmacology of the dopamine-induced secretory response by using several dopamine receptor agonists and antagonists. The effects of dopamine can be mimicked by the agonists 6,7-ADTN and R(+)-Lisuride. Dopamine-induced secretions could be blocked by *cis*(Z)-flupentixol, chlorpromazine, and (+)-butaclamol. We performed PCR experiments with degenerate primers in order to clone dopamine and serotonin receptor genes from *Periplaneta.* So far, we obtained four PCR fragments with their sequences being similar to the *Drosophila* serotonin 5-HT1A, the *Drosophila* dopamine DopR2, the honeybee Am*tyr*1, and a non-characterized *Drosophila* biogenic amine receptor-gene, respectively. In addition, we have cloned a cDNA fragment coding for nitric oxide synthase (NOS). Using RT-PCR and NADPH diaphorase staining we proved NOS expression in the salivary glands. Dopamine-induced secretion is reduced by the NO donors sodium nitroprusside and DEA-NONOate. The effects of both substances were dose-dependent and reversible.

Molecular and pharmacological characterization of honeybee biogenic amine receptors

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Biogenic amines and their receptors are important regulators of physiological and behavioral functions in insects. So far we have cloned the cDNAs encoding a dopamine D1 receptor (AmDOP1), a tyramine receptor (AmTYR1), an octopamine receptor (AmOA1) and a putative serotonin receptor from the honeybee brain. In the honeybee, octopamine acts as a neuromodulator and is involved in learning and memory formation. The AmOA1 receptor is most closely related to octopamine receptors from *Drosophila melanogaster* and the pond snail, *Lymnea stagnalis*. Signaling properties of the cloned receptor were studied in transiently transfected HEK 293 cells. Nanomolar to micromolar concentrations of octopamine induced oscillating increases in the intracellular Ca^{2+} concentration. The Am*oa*1 gene is abundantly expressed in many somata of the honeybee brain, suggesting that this octopamine receptor is involved in the processing of sensory inputs, antennal motor outputs, and in higher-order brain functions. Recently, we have isolated the cDNA of a putative serotonin receptor from *Apis mellifera*. The deduced amino acid sequence of this honeybee receptor shows greatest sequence homology to 5-HT7 receptors from *Drosophila melanogaster* and *Aedes aegypti* as well as to 5-HT1A receptors from vertebrates. The functional properties of this receptor and its tissue distribution are currently under investigation.

Modulation of tyramine signaling by osmolality in the *Drosophila* malpighian tubule

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The biogenic amine tyramine acts as a diuretic agent in the Malpighian tubules of *Drosophila melanogaster*, boosting urine production by increasing the transepithelial chloride conductance. The effects of tyramine can be measured electrically as a depolarization of the transepithelial potential. The response of isolated tubules to tyramine can be modulated by changes in the osmolality of the peritubular bath; an increase in osmolality results in a decreased response to tyramine, while a decrease in osmolality results in an enhanced tyramine response. Because the tyraminergic pathway is likely to be constitutively activated *in vivo*, this modulation may allow for greater responsiveness of the tubule to changes in hemolymph osmolality. In response to a sustained osmotic challenge, the modulation of the tyramine response is not constant but adapts, such that the amplitude of the tyramine response returns towards the control value over the course of 5-10 minutes. Interestingly, in one strain of flies this adaptation does not occur. These flies are also salt-sensitive, dying on food containing 0.4M NaCl. Genetic studies are now underway to determine if both of these phenotypes are due to the same locus and to identify the affected gene. Supported by NIH R21DK60860 and Marquette University.

Rapid evolution of the butterfly eye through multiple independent duplications of the opsin gene family

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Lycaena butterflies have four major spectral classes of visual pigments in the compound eye photoreceptors with lambda-max values of 360 nm, 437 nm, 500 nm and 568 nm, respectively (Bernard and Remington 1991). In this study, we characterized these visual pigments molecularly. A cDNA library from the eyes of two male *Lycaena rubidus* was constructed and screened for opsin transcripts using 3' and 5' RACE (rapid amplification of cDNA ends), cloning, and automated sequencing. An exhaustive screen yielded four distinct full-length opsin mRNAs with varying lengths of 1738, 1500, 1514, 1521 bp, and which encode opsins of 381, 377, 380 and 382 amino acids. Phylogenetic analysis indicates that these four opsins cluster within clades of UV-, blue- and long wavelength (LW)-sensitive insect opsins, and are most closely related to their nymphalid homologues. Unexpectedly, we found that *L. rubidus* expresses duplicated blue-sensitive opsin genes in the retina. This is the first report of a duplication of the blue-sensitive opsin gene within insects. Secondly, this result suggests that the 500 nm visual pigment has likely evolved from a blue-sensitive ancestral pigment. This therefore represents the most red-shifted blue-sensitive opsin known in insects. Finally, these data indicate that compound eyes (and by extrapolation, color vision) has evolved rapidly between butterflies families with both the lycaenids and papilionids expressing the products of novel, independent opsin gene duplications.

Air flow sensing: from physical ecology in the field to nanotech implementation

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Air flow sensing mechanoreceptors on the cricket cerci are amongst the best known sensors in the animal kingdom, working at the limit of thermal noise. They enable crickets to perceive the slightest air movement generated by an approaching predator and to escape. This talk will review the advances of an EU project (IST - Future and Emerging Technologies) in bionics which aims at understanding the natural selection forces in the field and the biomechanical functioning of hairs as well as the integration into a hair canopy response. Our work aims furthermore at reproducing some of the findings at the micro/nanotechnological level as MEMS (micro-electro-mechanical systems) devices and to build a miniature cricket robot in which networks of living cells on silicon chips would be directly connected to the sensors. The talk will also highlight the unusual integrative approach this consortium has, from field ecology, material sciences & nanotechnology up to living computers, and will focus on the biological findings obtained so far. The Consortium is headed by J. Casas and his team, and is made of the teams of G. Krijnen (Micro- and Nanosensors- Twente), A. Offenhaüser (Surfaces and Interfaces - Jülich Forschungzentrum) and G. Jeronimidis (Material Sciences - Reading).

Neurotransmitter transporters in the cabbage looper moth (*Trichoplusia ni*)

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In the nervous system, neurotransmitter signaling molecules are taken back up after release into the synapse by specialized transport proteins located in the membranes of neurons and glial elements bordering the synaptic space. These proteins belong to widely divergent families present throughout eukaryotes and extending even to prokaryotes. We have characterized a group of neurotransmitter transporters from a pest insect, the cabbage looper *Trichoplusia ni*. These include transporters for most of the principal transmitter molecules in the insect nervous system; glutamate, GABA, dopamine, serotonin, octopamine, and a transporter for the acetylcholine precursor and breakdown product, choline. Most of the cabbage looper transporters correspond clearly to orthologous proteins in other insects as well as in other animals. One exception is a transporter we have identified with high affinity for octopamine. Genes for this moth transporter, which may represent an evolutionary counterpart of the vertebrate norepinephrine transporter, are not found in the completed dipteran genomes of either the fruit fly or mosquito. We have therefore compared the distribution of this transporter with those of other monoamine transporters in insects to assess its prevalence and also its potential as an insect or order-selective target for pest control.

Allometry of insect flight performance: the case of hovering orchid bees

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Limits to insect flight performance are difficult to evaluate because maximum capacity is both context-specific and difficult to identify unequivocally. Most generally, anatomical constraints on stroke amplitude and a systematic decline in wingbeat frequency with increasing body size suggest that the relative capacity for power production should increase in smaller insects. Use of low-density helium-oxygen mixtures and of a load-lifting method that imposes asymptotically increasing loads during ascent has permitted assessment of maximum performance in both hummingbirds and hymenopterans for the specific behavioural context of hovering flight. In orchid bees, load-lifting methods have been applied to twelve species with known phylogenetic relatedness to assess the allometry of force and power production within the subfamily. Vertical force production scales adversely with greater body mass even if flight muscle mass scales isometrically. These results are compared with existing theoretical estimates of the allometry of animal flight performance.

Molecular and pharmacological analysis of an octopamine receptor from American cockroach and fruit fly

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Octopamine regulates multiple physiological functions in invertebrates. The biological effects of octopamine and the pharmacology of octopamine receptors have been extensively studied in the American cockroach, *Periplaneta americana*. This study reports the cloning of the first octopamine receptor from *Periplaneta americana*. A cDNA encoding a putative 7 transmembrane receptor was isolated from the head of *Periplaneta americana*. The encoded protein contains 628 amino acids and has sequence similarity to other biogenic amine receptors. This protein was expressed in COS-7 cells for radioligand binding studies using the antagonist ^{3}H-yohimbine. Competitive binding comparing biogenic amines that could potentially function as endogenous ligands demonstrated this receptor had the highest affinity for octopamine (K_i=13.3 μM) followed by tyramine, dopamine, serotonin and histamine. Octopamine increased both cAMP levels (EC_{50}=1.62 μM) and intracellular concentrations of calcium through the receptor expressed in HEK-293 cells. Tyramine increased levels of both of these second messengers but only at significantly higher concentrations than octopamine. The cAMP increase by octopamine was independent of the increase in calcium. Competitive binding with antagonists revealed this receptor is similar to Lym oa_1 from *Lymnaea stagnalis*. The data indicate this cDNA is the first octopamine receptor cloned from *Periplaneta americana* and therefore has been named Pa oa_1.

Bio-inspired legged locomotion on land

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Running insects bounce from one tripod of legs to another. A single tripod functions as one leg in a human, two legs in a trotting dog or four legs in a trotting crab. All produce the same pattern of ground reaction forces that can be modelled by a mass atop a spring – much like a pogo stick. Remarkably, the relative stiffness of an insect's leg is similar to other runners that differ in skeletal type and posture. Dynamic testing of legs shows that they can be represented by a damped-spring. Insects run over rough terrain possessing obstacles three times their "hip" height without even decreasing their preferred speed. Dynamic models reveal that running insects can function as a passive, dynamic self-stabilizing system. Models respond to perturbations of the body by self-correcting without using sensory information from the environment. Tuned visco-elastic legs respond to perturbations *preflexively* – before reflexes. Perturbations of running insects reveal that recovery begins in less than 10 msec challenging the fastest known reflexes. New materials manufacturing techniques have resulted in bio-inspired tuned legs. The concepts of control discovered in insects have biologically inspired the fastest, most stable legged robots yet built.

The trans-membrane topology of an *Epiphyas postvittana* odourant receptor protein expressed using baculovirus in Sf9 cells

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Odourant receptor molecules in insects are thought to be G-protein receptors and hence members of the seven transmembrane protein super family of genes, characterised by having seven transmembrane domains but little or no sequence homology. However, the known insect odourant receptors have a number of conserved amino acids, particularly within the last third of the proteins that allow the alignment of the sequences and the assignment to a family. The candidate odourant receptor was mined from an *Epiphyas postvittana* antennal EST library and the sequence was extended by 5′ RACE. The green fluorescent protein gfp5 gene was translationally fused onto the 3′ end to track where in the insect cell the candidate receptor protein was targeted. A bona fide receptor protein would be targeted to the cell membrane where it would insert itself leaving its 3′ end inside the cell. A series of deletions were planned where the gfp was fused progressively further towards the 5′ end of the receptor based on an *in silico* model of the transmembrane domains. The fate of the gfp was determined with fluorescent anti-gfp antibodies which will bind to the undisrupted cells only if the gfp is on the outside of the cell membrane.

Structure and properties of hairy attachment devices

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Many animals bear leg attachment pads with an excellent ability to adhere to a smooth surface as well as to a variety of natural surfaces with rough profiles. There are two alternative designs of such systems: smooth and hairy. The smooth systems consist of soft deformable structures with a relatively smooth surface. Pads of geckos, flies, beetles, and spiders are covered by relatively long, deformable setae which, due to individual bending, increase the number of contacting points with the surface. Most recent data on hairy systems demonstrated their excellent adhesion and high defect tolerance of contact. The size of single points gets smaller and their density higher as the body mass increases. We provided the first experimental evidence of adhesion enhancement by division of contact area. A patterned surface, made out of polyvinylsiloxane (PVS), has significantly higher tenacity on a glass surface than a smooth sample made out of the same material. This effect is even more pronounced on curved substrata. An additional advantage of patterned surfaces is the reliability of contact on various surface profiles and the increased tolerance to defects of individual contacts.

Biomechanics of insect wing folding: a model for robotic applications

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Insect hind wings are densely folded to fit two requirements. Firstly they need to have a minimum size to be flight worthy, too small wings are not usable as aerofoils. Secondly, they need to fit in a small casing, which is usually formed by stiff fore wings. These are, however, small to ensure mobility of the insect, which has to find crevices or is swiftly hunting. To meet these specifications, wings are folded in a taxon-specific manner, and so Dermaptera (earwigs) and Coleoptera (beetles) were examined to detect general principles as well as ‚customized' solutions. Additionally, wings were compared with other folded structures and computer simulations. Insect wings use a remote and distributed iving system with muscles exclusively located in the thorax proper, and resilin in the wing span. In Dermaptera, wings are unfolded with the cerci while folding is driven by elasticity, whereas in Coleoptera, wings unfold by the muscles, and fold by abdomen action. Folding lines divide the wing span into a system of stiff areas, which do not follow the veins and work as a lever system. Resilin is used to prevent material fatigue and fracture. Insect wings are smart structures with functionality built into their design.

Colour pattern recognition in bees

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Colours are meaningful signals to bees, used to optimise their choices during foraging. In flowers they are often arranged in more or less complex patterns increasing the display variability between species. Little is known about which spectral and spatial cues are available to bees in floral patterns and how bees perceive and learn them. Behavioural studies revealed that achromatic visual cues determine the detectability of coloured patterns. Stimuli with bright surroundings and dim centres are detected from a greater distance than those with dim surroundings and bright centres. A quantitative analysis of natural floral displays evinces that plants exploit this visual ability. Chromatic cues seem to be mainly involved in pattern recognition. Bees evince two evaluation strategies for the colour elements of a learned concentric pattern which are related either to the area occupied by a colour or to a colour's salience correlating with the strength of chromatic contrast. In the latter case bees ignore obvious spatial cues while discriminating patterns. This perceptual salience is based on the properties of the bee's visual system, rather than its experience in the field.

Is *Papilio* color vision tetrachromatic?

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Papilio butterflies use color vision when searching for food. Their retina contains six types of spectral receptor; UV, violet, blue, green, red and broad-band. Do all of them contribute to color vision? To answer this question, we measured the discrimination ability of wavelength difference ($\Delta\lambda$ function) of foraging *Papilio.*
We trained female butterflies to take nectar on a certain wavelength of monochromatic light projected on a transparent screen from below. After 10 days of training we presented two monochromatic lights on the screen at the same time to the butterflies, one at the training wavelength and another at 0~10nm away from the training wavelength. We then let the butterflies select one of the lights by extending the proboscis towards it. It appeared that the butterflies are able to discriminate 5nm difference over the wavelength range from 400 to 600nm. In this range, there are high resolution regions at 430, 500 and 560nm. The behaviorally-determined $\Delta\lambda$ function well fitted to the predicted function with the assumption that UV, blue, green, and red receptors contribute to the behavioral task. Consequently we conclude that the *Papilio* butterfly has a tetrachromatic color vision system.

The aminergic control of reproductive tissues in the female locust, *Locusta migratoria***.**

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Successful fertilization of eggs in the female locust, *Locusta migratoria*, relies upon the coordination of spermathecal muscle movements with those of the oviducts during oviposition. We have examined the neurochemicals involved in the control of both spermathecal and oviducal muscle contractions. These neurochemicals include a variety of neuropeptides, but also involve amines, such as octopamine and tyramine. Dorsal unpaired median (DUM) neurons expressing octopamine-like and tyramine-like immunoreactivity are found within the penultimate abdominal ganglion with projections to the oviduct muscle. Octopamine-like immunoreactive DUMs in the last abdominal ganglion have been shown to project to the spermathecal muscle. Octopamine and tyramine are released by the innervation to the locust oviducts and inhibit contractions in a dose-dependent manner. The actions of octopamine are mediated by cyclic AMP whereas the mode of action of tyramine is more complex and may involve a variety of second messengers. In contrast, octopamine, acting via cyclic AMP, has stimulatory actions upon contractions of the spermatheca. The involvement of these amines in the control of reproductive tissues will be discussed.

Variation in eye structure in butterflies: how regional variation in acuity scales with body size

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Examining how a structure scales inter- and intraspecifically with body size can provide insight into how selection operates on that structure. Smaller compound eyes should have lower resolving power, as measured by interommatidial angle ($\Delta\varphi$) and facet diameter (*D*), than large eyes because diffraction becomes more severe as lens size decreases. In order to maximize acuity in important regions of the visual field, selection may drive small insects to sacrifice resolution in some parts of their eye so that acuity may be enhanced in other regions. To better understand how patterns of regional variation in visual acuity change with body size, we measured regional differences in $\Delta\varphi$ and *D* in twenty species of butterflies representing three families. We analyzed data at the level of species, family, and the dataset as a whole. We predicted that 1) visual acuity would increase with body size; however 2) smaller insects would have proportionally higher acuity. Furthermore, we predicted 3) that regional differences in acuity would be greater in smaller species. Data analysis revealed that the first two predictions held, while the third prediction did not. We examine outlier species and discuss possible reasons for their deviations from the general regression line.

Analysis of dopamine receptors from the honey bee, *Apis mellifera*

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Two dopamine receptor genes, *Amdop1* and *Amdop2* have been cloned from the honey bee (Blenau et al., 1998, J Neurochem 70:15-23; Humphries et al., 2003, J Neurobiol 55:315-330). Both receptors are D1-like in that they upregulate intracellular cAMP levels in the presence of dopamine, however, they exhibit distinctive functional and pharmacological properties (Mustard et al., 2003, Mol Brain Res 113: 67-77). Each receptor also shows an overlapping, but different, expression pattern in larval and adult honey bee brains (Kurshan, et al., 2003, J Comp Neurol 466: 91-103). Taken together, these results suggest that *Am*DOP1 and *Am*DOP2 receptors may play distinct roles in dopamine signaling. Both receptors are highly expressed in the mushroom bodies of worker honey bee brains. Mushroom bodies are believed to be important for learning and memory. To examine the roles of these dopamine receptors in olfactory learning, RNA interference (RNAi) was used to decrease the level of mRNA of each receptor in the mushroom bodies. Using the proboscis extension reflex, bees were then examined for changes in their ability to acquire and/or recall an association between an odor and a sucrose reward. Initial studies suggest that *Am*DOP1 and *Am*DOP2 may play distinct roles in olfactory learning.

Structural and conformational aspects of pyrokinin-like neuropeptide interaction with expressed receptors

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The interaction of several classes of pyrokinin-like neuropeptides with their respective expressed receptors is investigated from a structural and conformational perspective. The discussion concerns a broad class of insect neuropeptides that represent products of the Capa-gene. Structural and conformational aspects critical to the interaction of the pyrokinin/PBAN class of neuropeptides and another Capa-gene product, the ecdysis-triggering hormone (ETH), with respective, expressed lepidopteran receptors are described. These ligand-receptor interactions are compared with the structural and conformational requirements for diuretic activity of the CAP2b/PVK sub-class of pyrokinin-like neuropeptides, also of the broad Capa category, in a dipteran. Insect neuropeptides of the classes selected for this discussion are implicated in the regulation of critical processes in pest insects. However, they suffer from a relative lack of biostability and bioavailability that limits their effectiveness as tools for neuroendocrinologists and as pest management agents. Biostable/bioavailable mimetic analog development of insect neuropeptides will be facilitated by a through understanding of structural/conformational aspects of the interaction with their receptors.

Peptidergic signaling pathways in *Drosophila melanogaster*.

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Peptides play crucial roles as transmitters and modulators of numerous biological processes, thus, aberrant peptidergic signaling may result in slowed development or death. Components of regulatory and transduction pathways are targets for the design of a variety of molecules including pesticides, thus, it is important to elucidate mechanisms that underlie peptidergic signaling. An interdisciplinary approach in a scientifically versatile organism is a powerful way to identify molecular components of regulatory and transduction mechanisms. A precursor often contains several peptides, thus, it is important to know how the polyprotein is processed and when and where the gene products are released to elucidate regulatory and transduction mechanisms involved in peptidergic signaling. In the case of structurally-related peptides encoded in either a single precursor or multiple precursors, it is important to decipher structure-activity relationships to avoid ambiguous data or erroneous conclusions. An *in vivo* bioassay is useful to establish the structure-activity relationship of structurally similar peptides, elucidate molecular mechanisms involved in peptidergic signaling, and to suggest peptide function(s). Comparison of peptide structure-activity relationships in different tissue bioassays yields insight into specificity of signaling pathways. We investigate signaling using *Drosophila melanogaster* to delineate peptide transduction pathways and functions.

Eph-ephrin signals may mediate sorting of olfactory sensory axons in *Manduca sexta.*

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We are investigating the roles of the Eph family receptor tyrosine kinases and their ligand ephrins in the developing primary olfactory nerve pathway in the moth *Manduca Sexta* using a combination of molecular biological and cell culture experiments. *Manduca* expresses one Eph receptor (MsEph) and one ephrin ligand (MsEphrin). *In situ* labeling with Fc-fusion probes reveals that both Eph receptors and ephrins are expressed on axons of olfactory receptor cells (ORCs) during their ingrowth to the primary center, the antennal lobe (AL). Eph receptors and ephrins are differentially distributed among identifiable glomeruli such that glomeruli with high receptor staining show little or no ligand staining and vice versa, suggesting a complementary Eph/ephrin expression by subsets of ORC axons innervating a particular set of glomeruli. *In vitro*, ephrin-Fc and Eph-Fc, when present homogeneously in the substratum, inhibit neurite outgrowth from olfactory epithelial explants. In patterned substratum, neurites turn or stop upon encountering the test substratum. These observations indicate that MsEphrin can act as an inhibitor/repulsive cue for ORC axons. We hypothesize that Eph receptors and ephrins mediate axon sorting and fasciculation through repulsive axon-axon interactions. We are currently testing that hypothesis using RNAi to knockdown the expression of MsEph and MsEphrin.

Characterization of small motion detection neurons of *Eristalis tenax*

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The hoverfly *Eristalis tenax* exhibits typical syrphid behaviour, with stationary hovering followed by rapid pursuit of conspecifics. Male and (possibly) female flies depend on detection of small rapidly moving targets to find mates and defend their territory. Neurons specialized for detecting small targets against cluttered backgrounds can be found in the lobula complex (3rd optic ganglion) of the fly brain. We are characterizing the morphology and physiology of a range of lobula small target motion detecting (STMD) neurons using dye-fills and electrophysiological techniques. We define STMDs as neurons that give a powerful response to a small object (1-2 degrees square) moving across a bright or cluttered background, but weaker responses to larger size targets. We find a number of classes of STMDs with unique properties in both sexes. These include neurons with small excitatory receptive fields surrounded by distinctly shaped inhibitory areas, and wide-field STMDs that may be used for mate detection. STMDs generally give spiking responses, and have variable degrees of spontaneous spike activity, including neurons that are 'silent' until stimulated by a target. A prominent feature of some STMDs is the robustness of their responses in the presence of background motion.

What gives adaptive gain control in insect motion detection?

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The phenomenon of motion adaptation is widespread among animals, having been demonstrated in species as diverse as humans, wallabies and flies. Motion adaptation appears to have evolved to maximize efficiency of visual coding in neurons with limited signaling range. Recent work highlights controversy over the site and mechanism of adaptation. Hypotheses range from simple gain control in 'early' vision (contrast normalization), to adaptive feedback that alters the delay mechanisms within local motion detectors. We have shown that adaptation does not alter the velocity tuning of motion detectors, but instead induces large reductions in contrast gain. Using moving natural scenes and a probe-adapt-probe protocol, we show here that gain reduction is driven by a mechanism with broad velocity tuning. While the tuning of adaptation resembles that of motion detectors themselves (favoring a feedback hypothesis), its broadness suggests extremely high gain in the feedback pathway and permits powerful adaptation to stimuli that drive motion detectors weakly. This mechanism may optimize coding of image velocity independent of the contrast variations between different natural scenes. Our work suggests easily implemented elaborations to existing motion detector models that could impart similar robustness to artificial motion detectors based on the same principle.

Diuresis in the blood-feeding bug, ***Rhodnius prolixus*****: Involvement of serotonergic dorsal unpaired median neurons and peptidergic bilaterally paired neurons.**

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The blood-feeding bug, *Rhodnius prolixus*, ingests large blood meals followed by a period of rapid diuresis that is under neurohormonal control. We are interested in determining the identity of the neurohormones controlling diuresis, the timing of their release, their effect on the Malpighian tubules (and other tissues involved in diuresis) and their potential interactions. Within the mesothoracic ganglionic mass (MTGM) of *Rhodnius*, there exists a subpopulation of dorsal unpaired median (DUM) neurons that display serotonin-like immunoreactivity. These, and other DUMs, are also immunoreactive to a variety of insect neuropeptide families (e.g. allatostatin-like and Dippu DH-31-like peptides). Serotonin is a neurohormone in *Rhodnius*, released from neurohaemal sites formed from these serotonergic DUMs, that controls diuresis. We have also mapped the distribution within *Rhodnius* of neuropeptides shown to possess diuretic activity in other insects. Bilaterally-paired neurons of the MTGM display Locusta DP-like, kinin-like and Dippu DH-31-like immunoreactivity. The effects of these peptide families and serotonin on diuresis in *Rhodnius* will be discussed.

Colour vision in the European glow-worm, *Lampyris noctiluca*

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Traditionally North Americans enjoy twilight watching fireflies (Lampyridae) display. Species first seen after dusk glow yellowish-orange (peak 580nm), while nocturnal species glow green (550nm). Photoreceptor sensitivities match the bioluminescent spectrum. According to a classic account in visual ecology, this communication system is optimized for signal detection: long-wavelength signals of crepuscular species contrast against the leaf background, whilst at night the green signals maximize absolute sensitivity (H.H. Seliger et al. *Photochem. Photobiol.* **36**, 673, 1982). In Britain twilight is boring, with only the nocturnal glow-worm *Lampyris noctiluca*. Wingless females sit near the ground, and attract flying males with a leaf-green light. Male glow-worms have long been known to be insensitive to 480nm light. This insensitivity might be explained by coloured filter pigments. However, in our behavioural tests addition of blue light reduces the attractiveness of green light to male glow-worms. This means that they have colour vision, and implies that their phototaxis uses a chromatic (i.e. opponent mechanism). By comparison, the matched filtering account invokes a spectrally selective achromatic mechanism. Our observation presents a puzzle: why subtract a noisy short-wave receptor's response from a long-wave detector well matched to its task?

Responses of the silkworm tyramine receptor to ligand compounds

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The physiological study of tyramine lags behind those of octopamine and dopamine. The new discovery of ligands that selectively interact with the insect tyramine receptor would facilitate research into the tyraminergic signal transduction. To this end, we cloned B96Bom from the silkworm (*Bombyx mori*) and expressed it in HEK-293 cells. Tyramine attenuated forskolin-stimulated cAMP production by interacting with the B96Bom receptor more potently than octopamine and dopamine. Yohimbine abolished the tyramine attenuation of cAMP production. These findings indicate that the B96Bom receptor is a tyramine receptor. We synthesized phenyloxazoles and phenylethylamines to test their abilities as ligands for biogenic amine receptors. Most of the compounds showed positive and negative effects on cAMP production in the membrane preparation of the silkworm head. 5-(4-Hydroxyphenyl)oxazole (5PO4) outstandingly attenuated basal, octopamine-stimulated, and forskolin-stimulated cAMP production. Yohimbine, but not *R*(+)-SCH23390 and chlorpromazine, abolished the 5PO4 attenuation. However, 5PO4 failed to attenuate forskolin-stimulated cAMP production in the cloned tyramine receptor, although the compound bound to the receptor. Phenylethylamines showed higher affinity for the cloned tyramine receptor than 5PO4. Although 2-chloro-2-(4-chlorophenyl)ethylamine failed to attenuate forskolin-stimulated cAMP production, the compound abolished the tyramine attenuation of forskolin-stimulated cAMP production.

Leucokinin signal transduction in blood feeding vectors of disease, the mosquito *Aedes aegypti* **(L.) and the southern cattle tick,** *Boophilus microplus* **(Canestrini).**

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Two different blood-feeding arthropods serve the study of leucokinin signal transduction and its relationship to diuresis and water balance. In the mosquito *Aedes aegypti*, the three *Aedes* leucokinin peptides stimulate total urine production in a dose-dependent manner *in vivo* (Cady and Hagedorn, 1999). In contrast, the specific function of myokinins in ticks is unknown and none have yet been isolated from this group of arthropods. We have cloned and functionally characterized a myokinin receptor from the Southern cattle tick (Holmes et al., 2003) and also identified its likely ortholog in the female of *Aedes aegypti*. Tissue localization of the receptors in these vectors was achieved through RT-PCR and/or *in situ* hybridization techniques. Receptor functionality and ligand specificity was analyzed by expressing the receptors in mammalian CHO-K1 cells followed by challenge with peptide agonists and measurements of intracellular calcium by fluorescence cytometry. In this system, the receptors are functional at physiological leucokinin concentrations. Differences in leucokinin signal transduction between *Aedes aegypti* and *Drosophila melanogaster* will be discussed.

Understanding serotonin signal transduction in arthropods vectors of disease: *Aedes aegypti* **and** *Boophilus microplus*

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In the mosquito *Aedes aegypti*, serotonin (5-HT) changes the rhythm of contractions in the female hindgut and increases fluid secretion in the larval Malpighian tubule. We investigated the potential role of 5-HT as a diuretic hormone in adults. For this we cloned a serotonin receptor cDNA (from female *A. aegypti* Malpighian tubule) that is similar to the 5-HT$_7$ receptor from *Drosophila melanogaster*. The transcript was localized in the tracheolar cells associated with the Malpighian tubules but no signal was detectable in the tubule epithelium. Immunohistochemistry with specific antibodies confirmed the receptor expression in tracheolar cells and hindgut, and western blots of these tissues showed the expected 50kDa band. The results suggest a role for serotonin in respiration. Functional expression of the *Aedes* 5-HT$_7$-like receptor in CHO-K1 cells showed that the receptor positively coupled to a Gs protein, increasing intracellular cAMP in response to 5-HT. Only 5-HT but not octopamine, dopamine or tyramine, induced cAMP. Agonists of the mammalian 5-HT$_7$ receptor had the following order of potency with the *Aedes* receptor: 5-HT >> 5-CT = 8-OH-DPAT >> pimozide. This is the first report on the functional expression of a mosquito neurohormone receptor. The characterization of a tick 5-HT receptor will be presented.

Honeybee vision and navigation: role of associative learning and cross-modal recall

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Insects learn and process environmental cues to orientate in their surroundings. The honeybee, *Apis mellifera*, flies rapidly and accurately to food sources that are often kilometres away. This is achieved by learning visual cues based on the distance and direction of the goal, the landmarks encountered en route, and the shape and colour of the desired flowers. Honeybees also learn chemical cues associated with food sources, such as the scent and the taste of the nectar. We investigated how the honeybee's capacity for associative learning and cross-modal recall assists her in navigating back to a known food source. We find that honeybees, trained to visit a scented sugar feeder placed at a specific outdoor location, can be induced to visit the same location simply by blowing the same scent into the hive, even when the destination no longer carries the food or the scent. Bees can be triggered in this way to fly to a specific location, or to find a specific colour. Our findings suggest that familiar nectar scents brought back into the hive by returning foragers, can induce recall of specific visual and route memories in experienced honeybees, and thus expedite navigation to the food source.

On moth's odorant receptors

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Odorant receptors (ORs) are quite essential in chemical communication between and among insects and their environment. Therefore ORs in Noctids were focused in foregoing study. We compared different ORs in antenna of 0-7 days old males and females of *Heliothis zea*. Additionaly the tissue distribution of ORs has been done in different parts of the insect's body: wings, legs, abdomens, thoraces and heads (animals between 0-4 days old). *Agrotis segetum* ORs sequences in antenna have been obtained as well. Following study has been performed using molecular techniques. Various pairs of primers were designed and PCRs have been performed followed by cloning and sequencing the corresponding PCR products. Obtained sequences were analysed by BioEdit computer programme.

PTTH signal transduction in the prothoracic gland: a tangled web with one purpose?

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Molting and metamorphosis in insects is controlled by several hormones, with 20-hydroxyecdyone (molting hormone) playing an overarching role. Prothoracicotropic hormone (PTTH), the best-understood regulator of ecdysteroidogenesis, stimulates ecdysteroid synthesis by the prothoracic glands. Work in the 1980s and 1990s revealed that PTTH activated a signal transduction cascade that included Ca^{2+}- influx, cAMP generation, phosphorylation of the ribosomal protein S6 and protein translation. In this talk, I will review more recent work, from our group and others, indicating that the PTTH signal transduction cascade is surprisingly complex. We now know that additional kinases are activated, including an extracellular signal-regulated kinase (ERK), a protein kinase C, and one or more unidentified tyrosine kinases. Ongoing studies also suggest PTTH-stimulated Ca^{2+}- influx is complexly regulated, with increases in cytoplasmic free Ca^{2+} coming from at least three sources, and that phospholipase C activation is among the earliest intracellular events triggered by PTTH. The composite data suggest that (1) PTTH regulates more processes in the prothoracic gland than ecdysteroid synthesis; (2) the study of phosphorylated proteins offers an avenue to identify these additional functions of PTTH; and (3) the control of ecdysteroid synthesis is subject to a Pandora's box of developmentally varying checks and balances.

Candidate Genes for Behavioral QTL in the Honeybee, *Apis mellifera.*

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The Honeybee Genome Project is providing an unprecedented opportunity for identifying and characterizing genes involved in the behaviour and brain function of social organisms. The honeybee genome has an unusually high recombination rate compared to those of other higher eukaryotic organisms. This high recombination rate aids in pinpointing genes because linkage maps rely on crossovers to determine position of markers. To take full advantage of this phenomenon, we have constructed a high-density genetic linkage map of the honeybee that contains over 1000 AFLP, RAPD microsatellite markers and sequence tagged sites (STS). There are more than 315 sequence tags on the genetic linkage map, sourced from sequenced AFLP, RAPD and microsatellite DNA fragments. This genetic map was constructed from a cross between a defensive African-derived strain of honeybee and a "gentle" strain of European honeybee and has allowed us to investigate QTL for defensive behaviours. These QTL regions have produced an intriguing list of candidate genes that may influence stinging and guarding behaviours. We hypothesise that natural selection has acted on genes that influence neuromodulator levels, hormone signaling and the visual system to produce more defensive genotypes of bees.

The role of protein phosphorylation in regulating the function of ecdysone receptor complex

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Insect signal transduction systems: current knowledge and future directions

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In biological systems, physiological responses to extracellular signals are elicited by activation of specific receptor proteins. This activation induces cellular effects via a process, generally designated as 'signal transduction'. Interestingly, the main categories of signal transducing receptor proteins are conserved in Arthropods. Moreover, this is also the case for most families of proteins that are components of intracellular signaling pathways. The recent technological revolution in genomics will certainly have an important impact on future signal transduction research. Convenient expression systems and functional assay procedures will be needed for studying the functional properties of the encoded predicted proteins. In this symposium, a number of conserved signaling protein families will be discussed, as well as recent advances and future directions in the analysis of their structural and functional properties. In more detail, we will review the current knowledge concerning the superfamily of G protein-coupled receptors. The study of evolutionary aspects of cellular signaling processes will probably reveal insect-specific features. Future challenges are to study the regulation, function and interplay of many different genes and to analyze the *in vivo* dynamics of functional interactions in biological systems.

Pieris rapae red and deep-red receptors use a green rhodopsin and two red filter types

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The visual pigments and spectral sensitivities of the long-wavelength photoreceptors of the small white butterfly *Pieris rapae* were studied by combined electrophysiology, molecular biology and optical modeling. *Pieris rapae* appears to have only one long-wavelength rhodopsin, PrL, which is expressed in the two distal photoreceptor cells (R3 and R4) as well as in the proximal photoreceptors (R5-8) in all three types of ommatidia of the *Pieris* fronto-ventral eye. The spectral sensitivities of R3 and R4 well approximate the absorption spectrum of a visual pigment with absorption maximum at 563 nm. The spectral sensitivities of R5-8 photoreceptors in ommatidial type I and III peak at 620 nm and those in type II ommatidia peak at 640 nm. The large shifts of the spectral sensitivities of the R5-8 photoreceptors with respect to the absorption spectrum of their visual pigment, R563, can be explained with the spectral filtering by pale-red (PR) and deep-red () screening pigments that are concentrated in clusters of granules near the rhabdom boundary. The peak absorbance of the two spectral filters appears to be approximately 1 (PR) and 2 ().

Behavioural and optical resolution of the honeybee eye.

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We compared behavioural resolution of the bee eye with the predictions of the optical model. The model assumed that bees failed to detect stimuli whose borders could not be resolved. The resolution of border detectors was estimated from the optics of the honeybee frontal eye. Bees were trained to detect oval stimuli whose diameter in a longitudinal direction was 4 times longer than in a short one. The ovals were either horizontally or vertically oriented. The optical model of border detection predicted the limiting visual angle for such ovals to be 9.5^0 (long diameter) for horizontally oriented ovals and 7.7^0 for vertically orientated ones. In the case of horizontal orientation bees could detect an oval subtending 11^0 (85% correct choices) but they fail to detect one subtending 7.6^0 (51% correct choices). In the case of vertical stimuli bees easily detected an oval subtending 11^0 (91% correct), the 7.6^0 oval was difficult to detect (69% correct) and they could not detect an oval subtending 6.4^0 (53% correct). Thus the results of behavioural experiments agree with the results of the model predictions, indicating that the resolution of the honeybee vision is close to the limit set by the optics of their eyes.

Nocturnal vision and landmark orientation in a tropical halictid bee

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Some bees and wasps have evolved nocturnal behaviour, presumably to exploit night-flowering plants or avoid predators. They display impressive visual behaviour despite having apposition compound eyes, a design adapted for bright light. In theory, this eye design should render these insects blind by mid-dusk. How then are they able to see at night? We studied female tropical nocturnal sweat bees (*Megalopta genalis*) and discovered that they are able to learn landmarks around their nest entrance prior to nocturnal foraging trips, and to use them to locate the nest upon return. This is the first evidence of nocturnal landmark orientation in insects. The morphology and optics of the eye, and the physiological properties of the photoreceptors, have evolved to give *Megalopta's* eyes almost 30 times greater sensitivity to light than the eyes of diurnal honeybees, but this alone is not sufficient to explain their nocturnal visual behaviour. This implies that sensitivity is improved by a strategy of photon summation in time and in space, the latter of which requires the presence of specialised cells that laterally connect ommatidia into groups. Cells absent in diurnal bees have now been identified in the lamina of *Megalopta's* optic lobe that may subserve spatial summation.

The mechanism of the flight motor given respiratory gas exchange in hawkmoths.

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Intratracheal pressure, O_2-content and CO_2-emission during tethered flight were analyzed at the anterior spiracles and mesortergal air sacs in hawkmoths using electronic pressure sensors, O_2 sensitive optodes and an URAS gas analyzer with split specimen chamber.Electrophysiological measurement of the flight muscles and of a hithertoo neglected special ventilatory muscle contribute to the understanding of the flow through mechanism. Deformations of the lateral thorax and their effect upon the spiracles were recorded under stroboscopic light. During shivering, ventilation pulses are generated by the flight muscles reminiscent of the known autoventilation mechanism with tidal air flow and the intratracheal O_2 content decreases astically. During steady flight, however, a unidirectional airstream is produced with a mean subatmospheric pressure at the first (mesothoracic) spiracles and a mean positive pressure in the mesotergal air sacs resulting in an increasing intratracheal O_2 content. Due to this pressure decrement during flight, CO_2 leaves only at the posterior spiracles. The suction force for the inspiration flow at the anterior spiracles is generated by the flight apparatus and a special ventilatory muscle under prevention of inspiration through the posterior thoracic spiracles. The result is an efficient separation of the CO_2 emission route from the inspiratory air stream.

Robot models of cricket behaviour

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Insects demonstrate flexible and adaptable sensory-motor control despite their relatively small nervous systems, suggesting efficient solutions to problems in robotics. Moreover, robots can be used to test our understanding of the mechanisms of animal behaviour, by implementing hypotheses and testing their capabilities in realistic situations. We have built a series of robots based on cricket phonotaxis, that is, the ability of female crickets to locate a mate by moving towards the auditory stimulus of the calling male. Three main aspects of recent work will be described. The first is the use of an outdoor robot, with locomotion inspired by insect walking, to test a spiking neural network mechanism for phonotaxis in natualistic conditions. The second is the integration of additional sensory systems, specifically the optomotor response, onto the robot model. It is shown that a number of different algorithms for combining these behaviours seem to produce similar behaviour in a simulation of cricket experiments. However the algorithms may be distinguished when we consider issues of the different dynamics of the real sensory systems on the robot, and of the plausibility of neural implementation for the different schemes. Third, current collaborative work is aimed at revising and refining the model through parallel behavioural, neurophysiological and modelling investigations on the cricket and the robot. This has led to a number of novel discoveries about the functioning of the biological system.

The curious engineering of the insect flight skeleton

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Insects are unique among flying machines in their use of bending and buckling of thin plates and shells, reinforced where necessary by ridges and tubes, to translate the cyclic contractions of the muscles into the complex, varying kinematics of the wings. While fine adjustments to the form of the wing stroke cycle need to be actively achieved, as much information as possible is stored in the morphology of the skeleton itself: in the disposition of rigid and flexible components in the pterothorax and wings. The degree and nature of rigidity and pliancy are influenced by structure at all levels of scale. Relevant structural variables include the arrangement of apodemes, sutures and sulci in the thorax, of the veins, ridges, relief and flexion-lines in the wings; and the local thickness, microstructural relief, ultrastructure and chemistry of the cuticle. Detailed morphology, manipulation, mechanical testing, and physical, analytical and mathematical modelling are revealing the subtleties of these fascinating, highly automated systems.

The honeybee is able to categorise natural visual images

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Recent work has revealed that monkeys as well as pigeons are able to categorise complex visual objects**.** We show here that the ability to group similar natural, visual images together extends to an invertebrate – the honeybee. Bees can be trained to distinguish between different types of naturally occurring scenes in a rather general way, and to group them into four distinct categories: landscapes, plant stems and two different kinds of flowers. They exhibit the same response to novel visual objects that differ greatly in their individual, low-level features, but belonging to one of the four categories. We exclude the possibility that they might be using single, low-level features alone as a cue to categorise these natural visual images and suggest that the categorisation is based on a combination of low-level features and configurational cues.

The visual ecology of hawkmoths

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The diurnal *Macroglossum stellatarum* and the nocturnal *Deilephila elpenor* are active in amatically different light intensities but both have colour vision. *D. elpenor* can discriminate colours in light intensities a million times weaker than the light intensities normal for *M. stellatarum*. Both species have colour constancy, which allows them to discriminate colours irrespectively of the spectrum of the illumination. This ability is explained by chromatic receptor adaptation according to the von Kries coefficient law. *M. stellatarum* learns the size, patterns and brightness of flowers. Both species learn the spatial location of rewarding stimuli, when they are difficult to distinguish by colour alone. Nocturnal sphingids mostly use odour to localize nectar flowers. However, in the day-active *M. stellatarum*, vision has become the dominating modality. When faced with a colour and an odour stimulus, naive *M. stellatarum* select the colour. The behaviour of flower-naive *D. elpenor* is the opposite. We are currently investigating the role of multi-modal interaction in learning experiments with both species.

Technical perfection in the sensory periphery: spider mechanoreception

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Despite the small size of their brain arthropods show complex and highly refined behavior. One of the foundations making this possible is the refinement and filter action of their sensory periphery. Spider mechanoreceptors exemplify this nicely. Being interfaces between environment and behavior they not only reflect details of the physical properties of the adequate stimulus but also characteristics of species-specific behavior. This will be illustrated taking two different types of sensory hairs as examples. (i) *Trichobothria* are *medium flow sensors* of outstanding sensitivity which is largely due to an intriguingly well "designed" interaction between the moving air and the hair. Using fluid mechanics this interaction can be modelled quantitatively and the hair´s mechanical and nervous responses related to prey catching behavior. (ii) *Tactile hairs* follow mechanical principles different from those of a medium flow sensor as seen from their morphology and mechanical properties and revealed by finite element analysis and micromechanical measurements. Bringing together neurobiology and engineering not only deepens our understanding of sensory systems but also helps in developing new, biology-inspired technical sensors.

Molecular and pharmacological characterization of *Drosophila* octopamine receptors

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In invertebrates, the biogenic amine octopamine regulates physiological functions including locomotion, neuronal development, circadian rhythm, as well as learning and memory. Pharmacological investigations indicate that octopamine mediates these effects by activation of several G protein-coupled receptor types.
We have cloned two genes from *Drosophila melanogaster* that encode three functional octopamine receptors. The first gene (Dm*oa*1) codes for two polypeptides which are generated by alternative splicing. The proteins share identical N-terminal halves but differ in the C-terminal half. When stimulated with nanomolar concentrations of octopamine, both splice variants induce Ca^{2+} release from internal stores. At high concentrations of octopamine ($^{3}10^{-6}$ M) the receptors also cause cAMP production. The second gene (Dm*oa*2) encodes a single polypeptide that specifically causes a rise of intracellular cAMP. The EC_{50} value is ~5×10^{-8} M octopamine. Tyramine activates all three receptors but at ^{3}100 fold higher concentrations. The biogenic amines dopamine and serotonin are non-effective at all. Developmental expression of Dm*oa* genes was assessed by RT-PCR. Overlapping but not identical expression patterns were observed for the individual transcripts. Since native octopamine receptors induce either cAMP production or Ca^{2+} release, the two genes may encode the complete set of octopamine receptors that are expressed in *Drosophila*.

The imaging properties of the dragonfly median ocellus

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Current literature describes the ocelli of pterygote insects as average illumination detectors, which serve to provide attitude information for the maintenance of flight equilibrium. This view has arisen because of several prominent features of the ocelli, including; fields of view centred on the horizon, under-focussed optics and a high rate of convergence from photoreceptors to second-order neurons which output to regions of the nervous system involved in flight control. However, recent research has indicated that dragonfly ocelli possess unusual structural and optical properties that are inconsistent with this view of ocellar function. In particular, from hanging-op preparations of the dragonfly median ocellar lens it has been determined that the image formed by this lens lies close to the underlying retinal layer, that the resolution of the image is dependent upon the orientation of an object and that the image is split laterally into left and right halves. Preliminary investigations also indicate the presence of a gradient of refractive index in the median ocellar lens. The optical properties of this lens and their relevance to dragonfly behaviour will be discussed.

Stop and go with NO: nitric oxide and cyclic nucleotides regulate neuronal motility in an insect embryo

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The dynamic regulation of nitric oxide synthase (NOS) activity and cyclic GMP levels suggests a functional role in the development of nervous systems. Here, we report evidence for a key role of the NO/cGMP signalling cascade on migration of postmitotic neurons in the enteric nervous system of the embryonic grasshopper. During development, a population of enteric neurons migrates on the surface of the midgut. These midgut (MG) neurons exhibit nitric oxide-induced cGMP-immunoreactivity coinciding with the migratory phase. Using a histochemical marker for NOS, we identified potential sources of NO in subsets of the midgut cells below the migrating MG neurons. Inhibition of endogenous NOS, sGC, and protein kinase G (PKG) activity in whole embryo culture results in a significant retardation of MG neuron migration. This inhibition can be rescued by supplementing with protoporphyrin IX free acid an activator of sGC, and membrane-permeant cGMP, indicating that NO/cGMP signalling is crucial for MG neuron migration. Conversely, the stimulation of the cAMP/protein kinase A signalling cascade results in an inhibition of cell migration. Activation of either the cGMP or the cAMP cascade influences the cellular distribution of F-actin in neuronal somata in a complementary fashion. The cytochemical stainings and experimental manipulations of cyclic nucleotide levels provide clear evidence that NO/cGMP/PKG signalling is permissive for MG neuron migration whereas the cAMP/PKA cascade may be a negative regulator. Since NO/cGMP signaling is also involved in axonal extension of pioneer neurons, these investigations reveal an accessible insect model in which the role of the NO/cGMP/PKG signalling cascade on neuronal cell motility can be analyzed in a natural setting.

Excitation-secretion coupling in the salivary glands of *Periplaneta americana*

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The acinar salivary glands of the cockroach, *Periplaneta americana*, are innervated by dopaminergic and serotonergic nerve fibers. Serotonin stimulates the secretion of protein-rich saliva, whereas dopamine causes the production of protein-free saliva. This suggests that dopamine acts selectively on ion-transporting peripheral cells within the acini and the duct cells, and serotonin acts on the protein-producing central cells of the acini. We investigated the pharmacology of the dopamine-induced secretory response by using several dopamine receptor agonists and antagonists. The effects of dopamine can be mimicked by the agonists 6,7-ADTN and R(+)-Lisuride. Dopamine-induced secretions could be blocked by *cis*(Z)-flupentixol, chlorpromazine, and (+)-butaclamol. We performed PCR experiments with degenerate primers in order to clone dopamine and serotonin receptor genes from *Periplaneta.* So far, we obtained four PCR fragments with their sequences being similar to the *Drosophila* serotonin 5-HT1A, the *Drosophila* dopamine DopR2, the honeybee Am*tyr*1, and a non-characterized *Drosophila* biogenic amine receptor-gene, respectively. In addition, we have cloned a cDNA fragment coding for nitric oxide synthase (NOS). Using RT-PCR and NADPH diaphorase staining we proved NOS expression in the salivary glands. Dopamine-induced secretion is reduced by the NO donors sodium nitroprusside and DEA-NONOate. The effects of both substances were dose-dependent and reversible.

Molecular and pharmacological characterization of honeybee biogenic amine receptors

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Biogenic amines and their receptors are important regulators of physiological and behavioral functions in insects. So far we have cloned the cDNAs encoding a dopamine D1 receptor (AmDOP1), a tyramine receptor (AmTYR1), an octopamine receptor (AmOA1) and a putative serotonin receptor from the honeybee brain. In the honeybee, octopamine acts as a neuromodulator and is involved in learning and memory formation. The AmOA1 receptor is most closely related to octopamine receptors from *Drosophila melanogaster* and the pond snail, *Lymnea stagnalis*. Signaling properties of the cloned receptor were studied in transiently transfected HEK 293 cells. Nanomolar to micromolar concentrations of octopamine induced oscillating increases in the intracellular Ca^{2+} concentration. The Am*oa*1 gene is abundantly expressed in many somata of the honeybee brain, suggesting that this octopamine receptor is involved in the processing of sensory inputs, antennal motor outputs, and in higher-order brain functions. Recently, we have isolated the cDNA of a putative serotonin receptor from *Apis mellifera*. The deduced amino acid sequence of this honeybee receptor shows greatest sequence homology to 5-HT7 receptors from *Drosophila melanogaster* and *Aedes aegypti* as well as to 5-HT1A receptors from vertebrates. The functional properties of this receptor and its tissue distribution are currently under investigation.

Modulation of tyramine signaling by osmolality in the *Drosophila* malpighian tubule

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The biogenic amine tyramine acts as a diuretic agent in the Malpighian tubules of *Drosophila melanogaster*, boosting urine production by increasing the transepithelial chloride conductance. The effects of tyramine can be measured electrically as a depolarization of the transepithelial potential. The response of isolated tubules to tyramine can be modulated by changes in the osmolality of the peritubular bath; an increase in osmolality results in a decreased response to tyramine, while a decrease in osmolality results in an enhanced tyramine response. Because the tyraminergic pathway is likely to be constitutively activated *in vivo*, this modulation may allow for greater responsiveness of the tubule to changes in hemolymph osmolality. In response to a sustained osmotic challenge, the modulation of the tyramine response is not constant but adapts, such that the amplitude of the tyramine response returns towards the control value over the course of 5-10 minutes. Interestingly, in one strain of flies this adaptation does not occur. These flies are also salt-sensitive, dying on food containing 0.4M NaCl. Genetic studies are now underway to determine if both of these phenotypes are due to the same locus and to identify the affected gene. Supported by NIH R21DK60860 and Marquette University.

Rapid evolution of the butterfly eye through multiple independent duplications of the opsin gene family

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Lycaena butterflies have four major spectral classes of visual pigments in the compound eye photoreceptors with lambda-max values of 360 nm, 437 nm, 500 nm and 568 nm, respectively (Bernard and Remington 1991). In this study, we characterized these visual pigments molecularly. A cDNA library from the eyes of two male *Lycaena rubidus* was constructed and screened for opsin transcripts using 3’ and 5’ RACE (rapid amplification of cDNA ends), cloning, and automated sequencing. An exhaustive screen yielded four distinct full-length opsin mRNAs with varying lengths of 1738, 1500, 1514, 1521 bp, and which encode opsins of 381, 377, 380 and 382 amino acids. Phylogenetic analysis indicates that these four opsins cluster within clades of UV-, blue- and long wavelength (LW)-sensitive insect opsins, and are most closely related to their nymphalid homologues. Unexpectedly, we found that *L. rubidus* expresses duplicated blue-sensitive opsin genes in the retina. This is the first report of a duplication of the blue-sensitive opsin gene within insects. Secondly, this result suggests that the 500 nm visual pigment has likely evolved from a blue-sensitive ancestral pigment. This therefore represents the most red-shifted blue-sensitive opsin known in insects. Finally, these data indicate that compound eyes (and by extrapolation, color vision) has evolved rapidly between butterflies families with both the lycaenids and papilionids expressing the products of novel, independent opsin gene duplications.

Air flow sensing: from physical ecology in the field to nanotech implementation

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Air flow sensing mechanoreceptors on the cricket cerci are amongst the best known sensors in the animal kingdom, working at the limit of thermal noise. They enable crickets to perceive the slightest air movement generated by an approaching predator and to escape. This talk will review the advances of an EU project (IST - Future and Emerging Technologies) in bionics which aims at understanding the natural selection forces in the field and the biomechanical functioning of hairs as well as the integration into a hair canopy response. Our work aims furthermore at reproducing some of the findings at the micro/nanotechnological level as MEMS (micro-electro-mechanical systems) devices and to build a miniature cricket robot in which networks of living cells on silicon chips would be directly connected to the sensors. The talk will also highlight the unusual integrative approach this consortium has, from field ecology, material sciences & nanotechnology up to living computers, and will focus on the biological findings obtained so far. The Consortium is headed by J. Casas and his team, and is made of the teams of G. Krijnen (Micro- and Nanosensors- Twente), A. Offenhaüser (Surfaces and Interfaces - Jülich Forschungzentrum) and G. Jeronimidis (Material Sciences - Reading).

Neurotransmitter transporters in the cabbage looper moth (*Trichoplusia ni*)

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In the nervous system, neurotransmitter signaling molecules are taken back up after release into the synapse by specialized transport proteins located in the membranes of neurons and glial elements bordering the synaptic space. These proteins belong to widely divergent families present throughout eukaryotes and extending even to prokaryotes. We have characterized a group of neurotransmitter transporters from a pest insect, the cabbage looper *Trichoplusia ni*. These include transporters for most of the principal transmitter molecules in the insect nervous system; glutamate, GABA, dopamine, serotonin, octopamine, and a transporter for the acetylcholine precursor and breakdown product, choline. Most of the cabbage looper transporters correspond clearly to orthologous proteins in other insects as well as in other animals. One exception is a transporter we have identified with high affinity for octopamine. Genes for this moth transporter, which may represent an evolutionary counterpart of the vertebrate norepinephrine transporter, are not found in the completed dipteran genomes of either the fruit fly or mosquito. We have therefore compared the distribution of this transporter with those of other monoamine transporters in insects to assess its prevalence and also its potential as an insect or order-selective target for pest control.

Allometry of insect flight performance: the case of hovering orchid bees

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Limits to insect flight performance are difficult to evaluate because maximum capacity is both context-specific and difficult to identify unequivocally. Most generally, anatomical constraints on stroke amplitude and a systematic decline in wingbeat frequency with increasing body size suggest that the relative capacity for power production should increase in smaller insects. Use of low-density helium-oxygen mixtures and of a load-lifting method that imposes asymptotically increasing loads during ascent has permitted assessment of maximum performance in both hummingbirds and hymenopterans for the specific behavioural context of hovering flight. In orchid bees, load-lifting methods have been applied to twelve species with known phylogenetic relatedness to assess the allometry of force and power production within the subfamily. Vertical force production scales adversely with greater body mass even if flight muscle mass scales isometrically. These results are compared with existing theoretical estimates of the allometry of animal flight performance.

Molecular and pharmacological analysis of an octopamine receptor from American cockroach and fruit fly

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Octopamine regulates multiple physiological functions in invertebrates. The biological effects of octopamine and the pharmacology of octopamine receptors have been extensively studied in the American cockroach, *Periplaneta americana*. This study reports the cloning of the first octopamine receptor from *Periplaneta americana*. A cDNA encoding a putative 7 transmembrane receptor was isolated from the head of *Periplaneta americana*. The encoded protein contains 628 amino acids and has sequence similarity to other biogenic amine receptors. This protein was expressed in COS-7 cells for radioligand binding studies using the antagonist ^{3}H-yohimbine. Competitive binding comparing biogenic amines that could potentially function as endogenous ligands demonstrated this receptor had the highest affinity for octopamine (K_i=13.3 μM) followed by tyramine, dopamine, serotonin and histamine. Octopamine increased both cAMP levels (EC_{50}=1.62 μM) and intracellular concentrations of calcium through the receptor expressed in HEK-293 cells. Tyramine increased levels of both of these second messengers but only at significantly higher concentrations than octopamine. The cAMP increase by octopamine was independent of the increase in calcium. Competitive binding with antagonists revealed this receptor is similar to Lym oa_1 from *Lymnaea stagnalis*. The data indicate this cDNA is the first octopamine receptor cloned from *Periplaneta americana* and therefore has been named Pa oa_1.

Bio-inspired legged locomotion on land

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Running insects bounce from one tripod of legs to another. A single tripod functions as one leg in a human, two legs in a trotting dog or four legs in a trotting crab. All produce the same pattern of ground reaction forces that can be modelled by a mass atop a spring – much like a pogo stick. Remarkably, the relative stiffness of an insect's leg is similar to other runners that differ in skeletal type and posture. Dynamic testing of legs shows that they can be represented by a damped-spring. Insects run over rough terrain possessing obstacles three times their "hip" height without even decreasing their preferred speed. Dynamic models reveal that running insects can function as a passive, dynamic self-stabilizing system. Models respond to perturbations of the body by self-correcting without using sensory information from the environment. Tuned visco-elastic legs respond to perturbations *preflexively* – before reflexes. Perturbations of running insects reveal that recovery begins in less than 10 msec challenging the fastest known reflexes. New materials manufacturing techniques have resulted in bio-inspired tuned legs. The concepts of control discovered in insects have biologically inspired the fastest, most stable legged robots yet built.

The trans-membrane topology of an *Epiphyas postvittana* odourant receptor protein expressed using baculovirus in Sf9 cells

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Odourant receptor molecules in insects are thought to be G-protein receptors and hence members of the seven transmembrane protein super family of genes, characterised by having seven transmembrane domains but little or no sequence homology. However, the known insect odourant receptors have a number of conserved amino acids, particularly within the last third of the proteins that allow the alignment of the sequences and the assignment to a family. The candidate odourant receptor was mined from an *Epiphyas postvittana* antennal EST library and the sequence was extended by 5′ RACE. The green fluorescent protein gfp5 gene was translationally fused onto the 3′ end to track where in the insect cell the candidate receptor protein was targeted. A bona fide receptor protein would be targeted to the cell membrane where it would insert itself leaving its 3′ end inside the cell. A series of deletions were planned where the gfp was fused progressively further towards the 5′ end of the receptor based on an *in silico* model of the transmembrane domains. The fate of the gfp was determined with fluorescent anti-gfp antibodies which will bind to the undisrupted cells only if the gfp is on the outside of the cell membrane.

Structure and properties of hairy attachment devices

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Many animals bear leg attachment pads with an excellent ability to adhere to a smooth surface as well as to a variety of natural surfaces with rough profiles. There are two alternative designs of such systems: smooth and hairy. The smooth systems consist of soft deformable structures with a relatively smooth surface. Pads of geckos, flies, beetles, and spiders are covered by relatively long, deformable setae which, due to individual bending, increase the number of contacting points with the surface. Most recent data on hairy systems demonstrated their excellent adhesion and high defect tolerance of contact. The size of single points gets smaller and their density higher as the body mass increases. We provided the first experimental evidence of adhesion enhancement by division of contact area. A patterned surface, made out of polyvinylsiloxane (PVS), has significantly higher tenacity on a glass surface than a smooth sample made out of the same material. This effect is even more pronounced on curved substrata. An additional advantage of patterned surfaces is the reliability of contact on various surface profiles and the increased tolerance to defects of individual contacts.

Biomechanics of insect wing folding: a model for robotic applications

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Insect hind wings are densely folded to fit two requirements. Firstly they need to have a minimum size to be flight worthy, too small wings are not usable as aerofoils. Secondly, they need to fit in a small casing, which is usually formed by stiff fore wings. These are, however, small to ensure mobility of the insect, which has to find crevices or is swiftly hunting. To meet these specifications, wings are folded in a taxon-specific manner, and so Dermaptera (earwigs) and Coleoptera (beetles) were examined to detect general principles as well as ‚customized' solutions. Additionally, wings were compared with other folded structures and computer simulations. Insect wings use a remote and distributed iving system with muscles exclusively located in the thorax proper, and resilin in the wing span. In Dermaptera, wings are unfolded with the cerci while folding is driven by elasticity, whereas in Coleoptera, wings unfold by the muscles, and fold by abdomen action. Folding lines divide the wing span into a system of stiff areas, which do not follow the veins and work as a lever system. Resilin is used to prevent material fatigue and fracture. Insect wings are smart structures with functionality built into their design.

Colour pattern recognition in bees

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Colours are meaningful signals to bees, used to optimise their choices during foraging. In flowers they are often arranged in more or less complex patterns increasing the display variability between species. Little is known about which spectral and spatial cues are available to bees in floral patterns and how bees perceive and learn them. Behavioural studies revealed that achromatic visual cues determine the detectability of coloured patterns. Stimuli with bright surroundings and dim centres are detected from a greater distance than those with dim surroundings and bright centres. A quantitative analysis of natural floral displays evinces that plants exploit this visual ability. Chromatic cues seem to be mainly involved in pattern recognition. Bees evince two evaluation strategies for the colour elements of a learned concentric pattern which are related either to the area occupied by a colour or to a colour's salience correlating with the strength of chromatic contrast. In the latter case bees ignore obvious spatial cues while discriminating patterns. This perceptual salience is based on the properties of the bee's visual system, rather than its experience in the field.

Quantification of silk trails made by lepidopteran larvae.

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Lepidopteran silk is made up of fibrous proteins extruded by a spinneret located near the mouthparts. The silk produced by lepidopteran larvae is known to play a role in feeding, survival, shelter and dispersal. It is also suspected that silk plays an important role in the adherence of larvae to substrates. Most studies have described the function of lepidopteran silking behaviour in relation to shelter building and construction of pupal cocoons. Our aim was to investigate the role of Lepidopteran silk in foraging behaviours such as crawling and feeding using the larvae of *Helicoverpa armigera* Hübner (Lepidoptera: Noctuidae), a major lepidopteran pest of crops world wide. Given that silk is practically invisible to the naked eye we developed a method for visualising silk trails in early instars using a fluorescent stain and fluorescence microscopy. This was combined with timelapse digital video-recording to correlate larval behaviour and rate of movement with the type and density of silk laid down over the animal's path. Distinctive silking patterns occur in association with larval foraging. Studies were conducted on whole plant material as well as a variety of artificial surfaces, leading to the development of a suitable model substrate.

Is *Papilio* color vision tetrachromatic?

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Papilio butterflies use color vision when searching for food. Their retina contains six types of spectral receptor; UV, violet, blue, green, red and broad-band. Do all of them contribute to color vision? To answer this question, we measured the discrimination ability of wavelength difference ($\Delta\lambda$ function) of foraging *Papilio.*
We trained female butterflies to take nectar on a certain wavelength of monochromatic light projected on a transparent screen from below. After 10 days of training we presented two monochromatic lights on the screen at the same time to the butterflies, one at the training wavelength and another at 0~10nm away from the training wavelength. We then let the butterflies select one of the lights by extending the proboscis towards it. It appeared that the butterflies are able to discriminate 5nm difference over the wavelength range from 400 to 600nm. In this range, there are high resolution regions at 430, 500 and 560nm. The behaviorally-determined $\Delta\lambda$ function well fitted to the predicted function with the assumption that UV, blue, green, and red receptors contribute to the behavioral task. Consequently we conclude that the *Papilio* butterfly has a tetrachromatic color vision system.

The aminergic control of reproductive tissues in the female locust, *Locusta migratoria***.**

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Successful fertilization of eggs in the female locust, *Locusta migratoria*, relies upon the coordination of spermathecal muscle movements with those of the oviducts during oviposition. We have examined the neurochemicals involved in the control of both spermathecal and oviducal muscle contractions. These neurochemicals include a variety of neuropeptides, but also involve amines, such as octopamine and tyramine. Dorsal unpaired median (DUM) neurons expressing octopamine-like and tyramine-like immunoreactivity are found within the penultimate abdominal ganglion with projections to the oviduct muscle. Octopamine-like immunoreactive DUMs in the last abdominal ganglion have been shown to project to the spermathecal muscle. Octopamine and tyramine are released by the innervation to the locust oviducts and inhibit contractions in a dose-dependent manner. The actions of octopamine are mediated by cyclic AMP whereas the mode of action of tyramine is more complex and may involve a variety of second messengers. In contrast, octopamine, acting via cyclic AMP, has stimulatory actions upon contractions of the spermatheca. The involvement of these amines in the control of reproductive tissues will be discussed.

Variation in eye structure in butterflies: how regional variation in acuity scales with body size

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Examining how a structure scales inter- and intraspecifically with body size can provide insight into how selection operates on that structure. Smaller compound eyes should have lower resolving power, as measured by interommatidial angle ($\Delta\varphi$) and facet diameter (D), than large eyes because diffraction becomes more severe as lens size decreases. In order to maximize acuity in important regions of the visual field, selection may drive small insects to sacrifice resolution in some parts of their eye so that acuity may be enhanced in other regions. To better understand how patterns of regional variation in visual acuity change with body size, we measured regional differences in $\Delta\varphi$ and D in twenty species of butterflies representing three families. We analyzed data at the level of species, family, and the dataset as a whole. We predicted that 1) visual acuity would increase with body size; however 2) smaller insects would have proportionally higher acuity. Furthermore, we predicted 3) that regional differences in acuity would be greater in smaller species. Data analysis revealed that the first two predictions held, while the third prediction did not. We examine outlier species and discuss possible reasons for their deviations from the general regression line.

Analysis of dopamine receptors from the honey bee, *Apis mellifera*

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Two dopamine receptor genes, *Amdop1* and *Amdop2* have been cloned from the honey bee (Blenau et al., 1998, J Neurochem 70:15-23; Humphries et al., 2003, J Neurobiol 55:315-330). Both receptors are D1-like in that they upregulate intracellular cAMP levels in the presence of dopamine, however, they exhibit distinctive functional and pharmacological properties (Mustard et al., 2003, Mol Brain Res 113: 67-77). Each receptor also shows an overlapping, but different, expression pattern in larval and adult honey bee brains (Kurshan, et al., 2003, J Comp Neurol 466: 91-103). Taken together, these results suggest that *Am*DOP1 and *Am*DOP2 receptors may play distinct roles in dopamine signaling. Both receptors are highly expressed in the mushroom bodies of worker honey bee brains. Mushroom bodies are believed to be important for learning and memory. To examine the roles of these dopamine receptors in olfactory learning, RNA interference (RNAi) was used to decrease the level of mRNA of each receptor in the mushroom bodies. Using the proboscis extension reflex, bees were then examined for changes in their ability to acquire and/or recall an association between an odor and a sucrose reward. Initial studies suggest that *Am*DOP1 and *Am*DOP2 may play distinct roles in olfactory learning.

Structural and conformational aspects of pyrokinin-like neuropeptide interaction with expressed receptors

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The interaction of several classes of pyrokinin-like neuropeptides with their respective expressed receptors is investigated from a structural and conformational perspective. The discussion concerns a broad class of insect neuropeptides that represent products of the Capa-gene. Structural and conformational aspects critical to the interaction of the pyrokinin/PBAN class of neuropeptides and another Capa-gene product, the ecdysis-triggering hormone (ETH), with respective, expressed lepidopteran receptors are described. These ligand-receptor interactions are compared with the structural and conformational requirements for diuretic activity of the CAP2b/PVK sub-class of pyrokinin-like neuropeptides, also of the broad Capa category, in a dipteran. Insect neuropeptides of the classes selected for this discussion are implicated in the regulation of critical processes in pest insects. However, they suffer from a relative lack of biostability and bioavailability that limits their effectiveness as tools for neuroendocrinologists and as pest management agents. Biostable/bioavailable mimetic analog development of insect neuropeptides will be facilitated by a through understanding of structural/conformational aspects of the interaction with their receptors.

Peptidergic signaling pathways in *Drosophila melanogaster***.**

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Peptides play crucial roles as transmitters and modulators of numerous biological processes, thus, aberrant peptidergic signaling may result in slowed development or death. Components of regulatory and transduction pathways are targets for the design of a variety of molecules including pesticides, thus, it is important to elucidate mechanisms that underlie peptidergic signaling. An interdisciplinary approach in a scientifically versatile organism is a powerful way to identify molecular components of regulatory and transduction mechanisms. A precursor often contains several peptides, thus, it is important to know how the polyprotein is processed and when and where the gene products are released to elucidate regulatory and transduction mechanisms involved in peptidergic signaling. In the case of structurally-related peptides encoded in either a single precursor or multiple precursors, it is important to decipher structure-activity relationships to avoid ambiguous data or erroneous conclusions. An *in vivo* bioassay is useful to establish the structure-activity relationship of structurally similar peptides, elucidate molecular mechanisms involved in peptidergic signaling, and to suggest peptide function(s). Comparison of peptide structure-activity relationships in different tissue bioassays yields insight into specificity of signaling pathways. We investigate signaling using *Drosophila melanogaster* to delineate peptide transduction pathways and functions.

Eph-ephrin signals may mediate sorting of olfactory sensory axons in *Manduca sexta*.

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We are investigating the roles of the Eph family receptor tyrosine kinases and their ligand ephrins in the developing primary olfactory nerve pathway in the moth *Manduca Sexta* using a combination of molecular biological and cell culture experiments. *Manduca* expresses one Eph receptor (MsEph) and one ephrin ligand (MsEphrin). *In situ* labeling with Fc-fusion probes reveals that both Eph receptors and ephrins are expressed on axons of olfactory receptor cells (ORCs) during their ingrowth to the primary center, the antennal lobe (AL). Eph receptors and ephrins are differentially distributed among identifiable glomeruli such that glomeruli with high receptor staining show little or no ligand staining and vice versa, suggesting a complementary Eph/ephrin expression by subsets of ORC axons innervating a particular set of glomeruli. *In vitro*, ephrin-Fc and Eph-Fc, when present homogeneously in the substratum, inhibit neurite outgrowth from olfactory epithelial explants. In patterned substratum, neurites turn or stop upon encountering the test substratum. These observations indicate that MsEphrin can act as an inhibitor/repulsive cue for ORC axons. We hypothesize that Eph receptors and ephrins mediate axon sorting and fasciculation through repulsive axon-axon interactions. We are currently testing that hypothesis using RNAi to knockdown the expression of MsEph and MsEphrin.

Characterization of small motion detection neurons of Eristalis tenax

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The hoverfly *Eristalis tenax* exhibits typical syrphid behaviour, with stationary hovering followed by rapid pursuit of conspecifics. Male and (possibly) female flies depend on detection of small rapidly moving targets to find mates and defend their territory. Neurons specialized for detecting small targets against cluttered backgrounds can be found in the lobula complex (3rd optic ganglion) of the fly brain. We are characterizing the morphology and physiology of a range of lobula small target motion detecting (STMD) neurons using dye-fills and electrophysiological techniques. We define STMDs as neurons that give a powerful response to a small object (1-2 degrees square) moving across a bright or cluttered background, but weaker responses to larger size targets. We find a number of classes of STMDs with unique properties in both sexes. These include neurons with small excitatory receptive fields surrounded by distinctly shaped inhibitory areas, and wide-field STMDs that may be used for mate detection. STMDs generally give spiking responses, and have variable degrees of spontaneous spike activity, including neurons that are 'silent' until stimulated by a target. A prominent feature of some STMDs is the robustness of their responses in the presence of background motion.

What gives adaptive gain control in insect motion detection?

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The phenomenon of motion adaptation is widespread among animals, having been demonstrated in species as diverse as humans, wallabies and flies. Motion adaptation appears to have evolved to maximize efficiency of visual coding in neurons with limited signaling range. Recent work highlights controversy over the site and mechanism of adaptation. Hypotheses range from simple gain control in 'early' vision (contrast normalization), to adaptive feedback that alters the delay mechanisms within local motion detectors. We have shown that adaptation does not alter the velocity tuning of motion detectors, but instead induces large reductions in contrast gain. Using moving natural scenes and a probe-adapt-probe protocol, we show here that gain reduction is driven by a mechanism with broad velocity tuning. While the tuning of adaptation resembles that of motion detectors themselves (favoring a feedback hypothesis), its broadness suggests extremely high gain in the feedback pathway and permits powerful adaptation to stimuli that drive motion detectors weakly. This mechanism may optimize coding of image velocity independent of the contrast variations between different natural scenes. Our work suggests easily implemented elaborations to existing motion detector models that could impart similar robustness to artificial motion detectors based on the same principle.

Diuresis in the blood-feeding bug, *Rhodnius prolixus*: Involvement of serotonergic dorsal unpaired median neurons and peptidergic bilaterally paired neurons.

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The blood-feeding bug, *Rhodnius prolixus*, ingests large blood meals followed by a period of rapid diuresis that is under neurohormonal control. We are interested in determining the identity of the neurohormones controlling diuresis, the timing of their release, their effect on the Malpighian tubules (and other tissues involved in diuresis) and their potential interactions. Within the mesothoracic ganglionic mass (MTGM) of *Rhodnius*, there exists a subpopulation of dorsal unpaired median (DUM) neurons that display serotonin-like immunoreactivity. These, and other DUMs, are also immunoreactive to a variety of insect neuropeptide families (e.g. allatostatin-like and Dippu DH-31-like peptides). Serotonin is a neurohormone in *Rhodnius*, released from neurohaemal sites formed from these serotonergic DUMs, that controls diuresis. We have also mapped the distribution within *Rhodnius* of neuropeptides shown to possess diuretic activity in other insects. Bilaterally-paired neurons of the MTGM display Locusta DP-like, kinin-like and Dippu DH-31-like immunoreactivity. The effects of these peptide families and serotonin on diuresis in *Rhodnius* will be discussed.

Colour vision in the European glow-worm, *Lampyris noctiluca*

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Traditionally North Americans enjoy twilight watching fireflies (Lampyridae) display. Species first seen after dusk glow yellowish-orange (peak 580nm), while nocturnal species glow green (550nm). Photoreceptor sensitivities match the bioluminescent spectrum. According to a classic account in visual ecology, this communication system is optimized for signal detection: long-wavelength signals of crepuscular species contrast against the leaf background, whilst at night the green signals maximize absolute sensitivity (H.H. Seliger et al. *Photochem. Photobiol.* **36**, 673, 1982). In Britain twilight is boring, with only the nocturnal glow-worm *Lampyris noctiluca.* Wingless females sit near the ground, and attract flying males with a leaf-green light. Male glow-worms have long been known to be insensitive to 480nm light. This insensitivity might be explained by coloured filter pigments. However, in our behavioural tests addition of blue light reduces the attractiveness of green light to male glow-worms. This means that they have colour vision, and implies that their phototaxis uses a chromatic (i.e. opponent mechanism). By comparison, the matched filtering account invokes a spectrally selective achromatic mechanism. Our observation presents a puzzle: why subtract a noisy short-wave receptor's response from a long-wave detector well matched to its task?

Responses of the silkworm tyramine receptor to ligand compounds

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The physiological study of tyramine lags behind those of octopamine and dopamine. The new discovery of ligands that selectively interact with the insect tyramine receptor would facilitate research into the tyraminergic signal transduction. To this end, we cloned B96Bom from the silkworm (*Bombyx mori*) and expressed it in HEK-293 cells. Tyramine attenuated forskolin-stimulated cAMP production by interacting with the B96Bom receptor more potently than octopamine and dopamine. Yohimbine abolished the tyramine attenuation of cAMP production. These findings indicate that the B96Bom receptor is a tyramine receptor. We synthesized phenyloxazoles and phenylethylamines to test their abilities as ligands for biogenic amine receptors. Most of the compounds showed positive and negative effects on cAMP production in the membrane preparation of the silkworm head. 5-(4-Hydroxyphenyl)oxazole (5PO4) outstandingly attenuated basal, octopamine-stimulated, and forskolin-stimulated cAMP production. Yohimbine, but not *R*(+)-SCH23390 and chlorpromazine, abolished the 5PO4 attenuation. However, 5PO4 failed to attenuate forskolin-stimulated cAMP production in the cloned tyramine receptor, although the compound bound to the receptor. Phenylethylamines showed higher affinity for the cloned tyramine receptor than 5PO4. Although 2-chloro-2-(4-chlorophenyl)ethylamine failed to attenuate forskolin-stimulated cAMP production, the compound abolished the tyramine attenuation of forskolin-stimulated cAMP production.

Leucokinin signal transduction in blood feeding vectors of disease, the mosquito *Aedes aegypti* (L.) and the southern cattle tick, *Boophilus microplus* (Canestrini).

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Two different blood-feeding arthropods serve the study of leucokinin signal transduction and its relationship to diuresis and water balance. In the mosquito *Aedes aegypti*, the three *Aedes* leucokinin peptides stimulate total urine production in a dose-dependent manner *in vivo* (Cady and Hagedorn, 1999). In contrast, the specific function of myokinins in ticks is unknown and none have yet been isolated from this group of arthropods. We have cloned and functionally characterized a myokinin receptor from the Southern cattle tick (Holmes et al., 2003) and also identified its likely ortholog in the female of *Aedes aegypti*. Tissue localization of the receptors in these vectors was achieved through RT-PCR and/or *in situ* hybridization techniques. Receptor functionality and ligand specificity was analyzed by expressing the receptors in mammalian CHO-K1 cells followed by challenge with peptide agonists and measurements of intracellular calcium by fluorescence cytometry. In this system, the receptors are functional at physiological leucokinin concentrations. Differences in leucokinin signal transduction between *Aedes aegypti* and *Drosophila melanogaster* will be discussed.

Understanding serotonin signal transduction in arthropods vectors of disease: *Aedes aegypti* **and** *Boophilus microplus*

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In the mosquito *Aedes aegypti*, serotonin (5-HT) changes the rhythm of contractions in the female hindgut and increases fluid secretion in the larval Malpighian tubule. We investigated the potential role of 5-HT as a diuretic hormone in adults. For this we cloned a serotonin receptor cDNA (from female *A. aegypti* Malpighian tubule) that is similar to the 5-HT$_7$ receptor from *Drosophila melanogaster*. The transcript was localized in the tracheolar cells associated with the Malpighian tubules but no signal was detectable in the tubule epithelium. Immunohistochemistry with specific antibodies confirmed the receptor expression in tracheolar cells and hindgut, and western blots of these tissues showed the expected 50kDa band. The results suggest a role for serotonin in respiration. Functional expression of the *Aedes* 5-HT$_7$-like receptor in CHO-K1 cells showed that the receptor positively coupled to a Gs protein, increasing intracellular cAMP in response to 5-HT. Only 5-HT but not octopamine, dopamine or tyramine, induced cAMP. Agonists of the mammalian 5-HT$_7$ receptor had the following order of potency with the *Aedes* receptor: 5-HT >> 5-CT = 8-OH-DPAT >> pimozide. This is the first report on the functional expression of a mosquito neurohormone receptor. The characterization of a tick 5-HT receptor will be presented.

Honeybee vision and navigation: role of associative learning and cross-modal recall

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Insects learn and process environmental cues to orientate in their surroundings. The honeybee, *Apis mellifera*, flies rapidly and accurately to food sources that are often kilometres away. This is achieved by learning visual cues based on the distance and direction of the goal, the landmarks encountered en route, and the shape and colour of the desired flowers. Honeybees also learn chemical cues associated with food sources, such as the scent and the taste of the nectar. We investigated how the honeybee's capacity for associative learning and cross-modal recall assists her in navigating back to a known food source. We find that honeybees, trained to visit a scented sugar feeder placed at a specific outdoor location, can be induced to visit the same location simply by blowing the same scent into the hive, even when the destination no longer carries the food or the scent. Bees can be triggered in this way to fly to a specific location, or to find a specific colour. Our findings suggest that familiar nectar scents brought back into the hive by returning foragers, can induce recall of specific visual and route memories in experienced honeybees, and thus expedite navigation to the food source.

On moth's odorant receptors

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Odorant receptors (ORs) are quite essential in chemical communication between and among insects and their environment. Therefore ORs in Noctids were focused in foregoing study. We compared different ORs in antenna of 0-7 days old males and females of *Heliothis zea*. Additionaly the tissue distribution of ORs has been done in different parts of the insect's body: wings, legs, abdomens, thoraces and heads (animals between 0-4 days old). *Agrotis segetum* ORs sequences in antenna have been obtained as well. Following study has been performed using molecular techniques. Various pairs of primers were designed and PCRs have been performed followed by cloning and sequencing the corresponding PCR products. Obtained sequences were analysed by BioEdit computer programme.

PTTH signal transduction in the prothoracic gland: a tangled web with one purpose?

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Molting and metamorphosis in insects is controlled by several hormones, with 20-hydroxyecdyone (molting hormone) playing an overarching role. Prothoracicotropic hormone (PTTH), the best-understood regulator of ecdysteroidogenesis, stimulates ecdysteroid synthesis by the prothoracic glands. Work in the 1980s and 1990s revealed that PTTH activated a signal transduction cascade that included Ca^{2+}- influx, cAMP generation, phosphorylation of the ribosomal protein S6 and protein translation. In this talk, I will review more recent work, from our group and others, indicating that the PTTH signal transduction cascade is surprisingly complex. We now know that additional kinases are activated, including an extracellular signal-regulated kinase (ERK), a protein kinase C, and one or more unidentified tyrosine kinases. Ongoing studies also suggest PTTH-stimulated Ca^{2+}- influx is complexly regulated, with increases in cytoplasmic free Ca^{2+} coming from at least three sources, and that phospholipase C activation is among the earliest intracellular events triggered by PTTH. The composite data suggest that (1) PTTH regulates more processes in the prothoracic gland than ecdysteroid synthesis; (2) the study of phosphorylated proteins offers an avenue to identify these additional functions of PTTH; and (3) the control of ecdysteroid synthesis is subject to a Pandora's box of developmentally varying checks and balances.

Candidate Genes for Behavioral QTL in the Honeybee, *Apis mellifera.*

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The Honeybee Genome Project is providing an unprecedented opportunity for identifying and characterizing genes involved in the behaviour and brain function of social organisms. The honeybee genome has an unusually high recombination rate compared to those of other higher eukaryotic organisms. This high recombination rate aids in pinpointing genes because linkage maps rely on crossovers to determine position of markers. To take full advantage of this phenomenon, we have constructed a high-density genetic linkage map of the honeybee that contains over 1000 AFLP, RAPD microsatellite markers and sequence tagged sites (STS). There are more than 315 sequence tags on the genetic linkage map, sourced from sequenced AFLP, RAPD and microsatellite DNA fragments. This genetic map was constructed from a cross between a defensive African-derived strain of honeybee and a "gentle" strain of European honeybee and has allowed us to investigate QTL for defensive behaviours. These QTL regions have produced an intriguing list of candidate genes that may influence stinging and guarding behaviours. We hypothesise that natural selection has acted on genes that influence neuromodulator levels, hormone signaling and the visual system to produce more defensive genotypes of bees.

The role of protein phosphorylation in regulating the function of ecdysone receptor complex

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Insect molting and metamorphosis are regulated by ecdysteroids. The action of ecdysteroids is mediated via ecdysone receptor (EcR) and its heterodimer, ultraspiracle protein (USP). Both EcR and USP are members of the nuclear receptor superfamily and ligand dependent transcriptional factors. Our recent data have shown that both EcR and USP are phosphoproteins and the expression and phosphorylation of EcR and USP are regulated by 20-hydroxyecdysone (20E). Phosphorylation has been shown to play a role in regulating the ligand-binding activity of the EcR/USP complex, presumably leading to the feedback regulation of ecdysteroid biosynthesis in the prothoracic gland of *Manduca sexta*. Protein kinase consensus recognition sequence analysis of *Drosophila* EcR and USP along with specific protein kinase inhibitor assays reveals that protein kinase (PKC), not casein kinase II (CKII), is involved in the EcR and USP phosphorylation. Our preliminary data indicate that inhibition of EcR and USP phosphorylation by specific PKC inhibitors blocks 20E-induced protein expression. The composite data suggest that protein phosphorylation plays an important role in regulating 20E-signaling pathway.

Insect signal transduction systems: current knowledge and future directions

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In biological systems, physiological responses to extracellular signals are elicited by activation of specific receptor proteins. This activation induces cellular effects via a process, generally designated as 'signal transduction'. Interestingly, the main categories of signal transducing receptor proteins are conserved in Arthropods. Moreover, this is also the case for most families of proteins that are components of intracellular signaling pathways. The recent technological revolution in genomics will certainly have an important impact on future signal transduction research. Convenient expression systems and functional assay procedures will be needed for studying the functional properties of the encoded predicted proteins. In this symposium, a number of conserved signaling protein families will be discussed, as well as recent advances and future directions in the analysis of their structural and functional properties. In more detail, we will review the current knowledge concerning the superfamily of G protein-coupled receptors. The study of evolutionary aspects of cellular signaling processes will probably reveal insect-specific features. Future challenges are to study the regulation, function and interplay of many different genes and to analyze the *in vivo* dynamics of functional interactions in biological systems.

Pieris rapae red and deep-red receptors use a green rhodopsin and two red filter types

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The visual pigments and spectral sensitivities of the long-wavelength photoreceptors of the small white butterfly *Pieris rapae* were studied by combined electrophysiology, molecular biology and optical modeling. *Pieris rapae* appears to have only one long-wavelength rhodopsin, PrL, which is expressed in the two distal photoreceptor cells (R3 and R4) as well as in the proximal photoreceptors (R5-8) in all three types of ommatidia of the *Pieris* fronto-ventral eye. The spectral sensitivities of R3 and R4 well approximate the absorption spectrum of a visual pigment with absorption maximum at 563 nm. The spectral sensitivities of R5-8 photoreceptors in ommatidial type I and III peak at 620 nm and those in type II ommatidia peak at 640 nm. The large shifts of the spectral sensitivities of the R5-8 photoreceptors with respect to the absorption spectrum of their visual pigment, R563, can be explained with the spectral filtering by pale-red (PR) and deep-red () screening pigments that are concentrated in clusters of granules near the rhabdom boundary. The peak absorbance of the two spectral filters appears to be approximately 1 (PR) and 2 ().

Behavioural and optical resolution of the honeybee eye.

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We compared behavioural resolution of the bee eye with the predictions of the optical model. The model assumed that bees failed to detect stimuli whose borders could not be resolved. The resolution of border detectors was estimated from the optics of the honeybee frontal eye. Bees were trained to detect oval stimuli whose diameter in a longitudinal direction was 4 times longer than in a short one. The ovals were either horizontally or vertically oriented. The optical model of border detection predicted the limiting visual angle for such ovals to be 9.5^0 (long diameter) for horizontally oriented ovals and 7.7^0 for vertically orientated ones. In the case of horizontal orientation bees could detect an oval subtending 11^0 (85% correct choices) but they fail to detect one subtending 7.6^0 (51% correct choices). In the case of vertical stimuli bees easily detected an oval subtending 11^0 (91% correct), the 7.6^0 oval was difficult to detect (69% correct) and they could not detect an oval subtending 6.4^0 (53% correct). Thus the results of behavioural experiments agree with the results of the model predictions, indicating that the resolution of the honeybee vision is close to the limit set by the optics of their eyes.

Nocturnal vision and landmark orientation in a tropical halictid bee

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Some bees and wasps have evolved nocturnal behaviour, presumably to exploit night-flowering plants or avoid predators. They display impressive visual behaviour despite having apposition compound eyes, a design adapted for bright light. In theory, this eye design should render these insects blind by mid-dusk. How then are they able to see at night? We studied female tropical nocturnal sweat bees (*Megalopta genalis*) and discovered that they are able to learn landmarks around their nest entrance prior to nocturnal foraging trips, and to use them to locate the nest upon return. This is the first evidence of nocturnal landmark orientation in insects. The morphology and optics of the eye, and the physiological properties of the photoreceptors, have evolved to give *Megalopta's* eyes almost 30 times greater sensitivity to light than the eyes of diurnal honeybees, but this alone is not sufficient to explain their nocturnal visual behaviour. This implies that sensitivity is improved by a strategy of photon summation in time and in space, the latter of which requires the presence of specialised cells that laterally connect ommatidia into groups. Cells absent in diurnal bees have now been identified in the lamina of *Megalopta's* optic lobe that may subserve spatial summation.

The mechanism of the flight motor given respiratory gas exchange in hawkmoths.

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Intratracheal pressure, O_2-content and CO_2-emission during tethered flight were analyzed at the anterior spiracles and mesortergal air sacs in hawkmoths using electronic pressure sensors, O_2 sensitive optodes and an URAS gas analyzer with split specimen chamber.Electrophysiological measurement of the flight muscles and of a hithertoo neglected special ventilatory muscle contribute to the understanding of the flow through mechanism. Deformations of the lateral thorax and their effect upon the spiracles were recorded under stroboscopic light. During shivering, ventilation pulses are generated by the flight muscles reminiscent of the known autoventilation mechanism with tidal air flow and the intratracheal O_2 content decreases astically. During steady flight, however, a unidirectional airstream is produced with a mean subatmospheric pressure at the first (mesothoracic) spiracles and a mean positive pressure in the mesotergal air sacs resulting in an increasing intratracheal O_2 content. Due to this pressure decrement during flight, CO_2 leaves only at the posterior spiracles. The suction force for the inspiration flow at the anterior spiracles is generated by the flight apparatus and a special ventilatory muscle under prevention of inspiration through the posterior thoracic spiracles. The result is an efficient separation of the CO_2 emission route from the inspiratory air stream.

Robot models of cricket behaviour

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Insects demonstrate flexible and adaptable sensory-motor control despite their relatively small nervous systems, suggesting efficient solutions to problems in robotics. Moreover, robots can be used to test our understanding of the mechanisms of animal behaviour, by implementing hypotheses and testing their capabilities in realistic situations. We have built a series of robots based on cricket phonotaxis, that is, the ability of female crickets to locate a mate by moving towards the auditory stimulus of the calling male. Three main aspects of recent work will be described. The first is the use of an outdoor robot, with locomotion inspired by insect walking, to test a spiking neural network mechanism for phonotaxis in natualistic conditions. The second is the integration of additional sensory systems, specifically the optomotor response, onto the robot model. It is shown that a number of different algorithms for combining these behaviours seem to produce similar behaviour in a simulation of cricket experiments. However the algorithms may be distinguished when we consider issues of the different dynamics of the real sensory systems on the robot, and of the plausibility of neural implementation for the different schemes. Third, current collaborative work is aimed at revising and refining the model through parallel behavioural, neurophysiological and modelling investigations on the cricket and the robot. This has led to a number of novel discoveries about the functioning of the biological system.

The curious engineering of the insect flight skeleton

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Insects are unique among flying machines in their use of bending and buckling of thin plates and shells, reinforced where necessary by ridges and tubes, to translate the cyclic contractions of the muscles into the complex, varying kinematics of the wings. While fine adjustments to the form of the wing stroke cycle need to be actively achieved, as much information as possible is stored in the morphology of the skeleton itself: in the disposition of rigid and flexible components in the pterothorax and wings. The degree and nature of rigidity and pliancy are influenced by structure at all levels of scale. Relevant structural variables include the arrangement of apodemes, sutures and sulci in the thorax, of the veins, ridges, relief and flexion-lines in the wings; and the local thickness, microstructural relief, ultrastructure and chemistry of the cuticle. Detailed morphology, manipulation, mechanical testing, and physical, analytical and mathematical modelling are revealing the subtleties of these fascinating, highly automated systems.

The honeybee is able to categorise natural visual images

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Recent work has revealed that monkeys as well as pigeons are able to categorise complex visual objects. We show here that the ability to group similar natural, visual images together extends to an invertebrate – the honeybee. Bees can be trained to distinguish between different types of naturally occurring scenes in a rather general way, and to group them into four distinct categories: landscapes, plant stems and two different kinds of flowers. They exhibit the same response to novel visual objects that differ greatly in their individual, low-level features, but belonging to one of the four categories. We exclude the possibility that they might be using single, low-level features alone as a cue to categorise these natural visual images and suggest that the categorisation is based on a combination of low-level features and configurational cues.

Insect visuomotor control & guidance & stabilisation of aerial robots.

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An evolutionary constraint linking innate immunity and development of the lepidopteran *Galleria mellonella*

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The impressive progress in *Drosophila* molecular genetics led to the discovery that homologues signalling pathways control both innate immunity and developmental processes in this insect. Toll-like signalling pathways mediate expression of genes encoding for antimicrobial peptides and those involved e.g in formation of the dorso-ventral polarity in *Drosophila*. Investigating molecular physiology of innate immunity and metamorphosis of the lepidopteran *Galleria mellonella* we identified a number of novel proteinase inhibitors that contribute to antimicrobial defence and developmental processes as well. Amazingly, we found that the vast majority of immune-related proteins are also induced upon release of non-regulated metalloproteinases in the haemolymph, independently whether they are of pathogen or endogenous origin. The presence of metalloproteinases in the haemolymph generates particular haemplymph protein fragments smaller than 3 kDa which represent potent activators of the Toll-pathway. The inability of the *G. mellonella* immune system to discriminate haemolymph proteins fragments produced by either pathogen-associated metalloproteinases or endogenous matrix metalloproteinases expressed during metamorphosis points to an evolutionary constraint that may explains involvement of Toll-like signalling pathways in distinct biological functions. As a hypothesis, this evolutionary constraint limits the physiological efficacy of insect innate immunity and influenced the coevolution of entomopathogens and their host insects.

Studies on food consumption and utilization of cabbage leaves by the larva of diamondback moth, *Plutella xylostella* **(L.) (Lepidoptera : Yponomeutidae).**

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In the North-Western Himalayan region cabbage is an important vegetable crop. Diamondback moth, *Plutella xylostella* is one of the several insect pests which causes damage to this crop. Studies on food consumption and utilization of cabbage leaves by the larva of the diamondback moth were carried out at 30^0C. The larva consumed 23.76 mg of fresh foliage of cabbage or 8.12 mg dry matter during its entire larval period, of which it passed out 5.34 mg (wet weight) or 1.78 mg (dry weight) as faecal matter. Food consumption also increased progressively, maximum being during IV instar (14.64 mg fresh weight or 4.94 mg dry matter). The maximum growth rate of 0.33 was observed during the II and IV instar. Maximum consumption index (CI) was recorded for the IV instar on fresh (3.09) as well as dry weight-fresh weight basis (1.04). Approximate digestibility (AD) was observed to be maximum during the IV instar (83.13% on fresh weight basis and 72.26% on dry weight-fresh weight basis). Efficiency of conversion of ingested/food (ECI) was maximum during the II instar on fresh weight basis (27.36%). On fresh weight basis, the efficiency of conversion of digested food (ECD) was maximum (47.27%) during the II instar.

An antimicrobial substance from the egg wax of the African bont tick *Amblyomma hebraeum.*

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Arthropods have a remarkable capacity to resist microbial infections. Although dead tick females tend to suffer microbial infestation after oviposition, the eggs next to them remain clean. When incubated with bacteria, tick (*A. hebraeum*) eggs inhibit the growth of *Escherichia coli* and *Serratia marcescens* (Gram-negative), but not the growth of *Staphylococcus epidermidis* and *Bacillus subtilis* (Gram-positive). Tick eggs are covered with a waterproofing material that also protects them against desiccation. This wax was extracted according to the Folch method. When incubated with bacteria in liquid cultures, the organic phase of the egg-wax extract appeared inactive, but the aqueous phase inhibited the growth of *S. epidermidis*. The aqueous phase was heat stable (100°C for 10 min), resistant to proteinase K treatment (15 min @ 55^0C) and has an amino acid concentration of ~1.1 mmol/mg. Egg wax is secreted by Géné's organ (a female accessory gland) during oviposition, and Géné's organ also appears to inhibit the growth of bacteria, suggesting that it may also secrete the antimicrobial substance.

Identification of cuticular specific protein with newly found motifs from the silkworm, *Bombyx mori*.

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Recent studies have shown that insect cuticle contains immune factors, such as prophenoloxidase cascade components, antimicrobial peptides, recognition proteins. To find new immune factors in insect cuticle, a primitive proteomic technic was performed. Silkworn larvae were treated with sandpaper and after 24 hours, cuticle proteins were extracted. The extract was subjected into SDS-PAGE and the electrophoregram was compared between injured larva and non-injured larva. As a result, a protein of about 130k Da was present only in injured cuticle. The band of this protein (cp130) was cut from the gel and treated with trypsin followed by LC-MS analysis. Several genes were listed as candidates for a gene encoding the cp130. Among them, a gene encoding 115k Da wing disk protein (*CB10*) was further investigated. Northern analysis resulted in that strong signal of *CB10* was observed in the epidermal cells of injured larva. This result suggested that *CB10* gene encodes cp130. The product of *CB10* had not been studied and its function remained unknown. The only characteristic of this protein is a presence of repeated motifs of 10 amino acids, which had never identified so far and may be identified for the first time in this study.

pH regulation and its physiology in the silk gland of *Bombyx mori*

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The silk gland of lepidopteran larvae produces vast amounts of silk proteins and stores them in the glandular lumen as liquid silk temporarily. We have explored the system regulating pH in the silk gland, because the gelation of fibroin is pH-dependent. By injecting the pH-sensitive dye (phenol red) into silkworm larvae, we have estimated the rough pH in the glandular lumen. In *B. mori*, the lumen of the middle silk gland (MSG: major reservoir of liquid silk) was acidic (~ pH 5) in the vigorous feeding period, which leads to gelation of silk proteins at MSG. When the larvae started spinning, the lumen in the MSG became neutral gradually. The acidification in the glandular lumen is established by the proton pump (V-ATPase) at the apical plasma membrane of MSG cells as well as that of the anterior silk gland (ASG) cells. Non-cocoon producing strain of *B. mori* (fibroin-deficient type) showed lesser acidification nature. Thus, the V-ATPase function seems to be linked to the presence of fibroin. Similar results were obtained in the silk gland of the wild silkworm, *Samia cynthia ricini*. The pH regulation in the silk gland seems to be common in the cocoon-producing larvae of Lepidoptera.

Immune-induced lebocin-like gene devoid of antibacterial lebocin peptide sequence from eri-silkworm, *Samia cynthia ricini*

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For more complete understanding of the insect immune responses, we have studied the genes that are differentially expressed in the fat body of *S. ricini* after bacterial challenge using suppression subtractive hybridization (SSH) method. One of the cDNA fragments from the subtracted cDNA library had a homology to lebocin gene from *Trichoplusia ni*, and the full length cDNA was obtained by 3' and 5'RACE methods. Lebocin is O-glycosylated proline-rich antibacterial peptide with 32 amino acid residues isolated from immunized hemolymph of *Bombyx mori*. *Bombyx* lebocin cDNA encoded precursor protein of 179 amino acid residues, from which the mature lebocin was thought to be cut out. The *Samia* lebocin-like protein with 162 amino acid residues has high homology with *Bombyx* and *Trichoplusia* lebocin in "prosegment" region, but no significant homology in the "mature" lebocin segment. Moreover, the segment of *Samia* corresponding to "mature" lebocin contained no O-glycosylation site and only 2 proline residues instead of 7 in *Bombyx*. The gene transcript was not detected in naive *Samia* larvae, but the expression was strongly induced by an injection of bacteria or peptidoglycan. The results suggest that the *Samia* gene does not code for antibacterial peptide, but an immune related protein of unknown function.

A respiratory hemocyanin from the stonefly *Perla marginata*

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Hemocyanins are large copper-proteins that serve for the transport of oxygen in many arthropod species. We have shown for the first time that hemocyanins also occur in insects. The hemocyanin of the stonefly *Perla marginata* is present at high concentrations the hemolymph of both nymphs and adults. It is a hexamer with of two distinct subunit types of 659 and 655 amino acids, respectively, which has a moderate oxygen affinity ($P_{50} \sim 8$ torr) and displays cooperative oxygen binding. No evidence was found for a hemocyanin in holometabolan insects, indicating that this type of respiratory protein was lost later in insect evolution. However, our results suggest that the evolution of the insect tracheal system was not accompanied by a loss of the respiratory pigment. In addition to the hemocyanin, *P. marginata* possess a hexamerin. Four distinct hexamerin cDNA sequences were obtained. Phylogenetic analyses show that the insect hemocyanins group with the insect hexamerins, both forming a common clade with the crustacean hemocyanins. The insect hemocyanins are paraphyletic in terms of the hexamerins, suggesting that these insect storage proteins evolved only from a particular hemocyanin subunit.

Characterisation of *Drosophila* hemoglobin: evidence for hemoglobin-mediated respiration in insects

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In contrast to previous assumptions probably most insects possess hemoglobins. The function of these respiratory proteins is still uncertain. We have characterised the hemoglobin of the fruitfly *Drosophila melanogaster*. It is a monomer of about 17 kDa that is not exported into the hemolymph. Recombinantly expressed *Drosophila*-hemoglobin exhibits a hexacoordinated oxygen-binding scheme, with an oxygen-affinity of 0.12 torr. Hemoglobin expression is mainly associated with the trachea and tracheoles, but is also present in the fat body cells surrounding the adult and larval organs. Embryos show additional expression in the yolk sac and vitellophages. The hemoglobin gene of *Drosophila* (*DmeGlob*1) is located on chromosome 3 at position 89A13-B1. Four different transcripts have been identified that use two distinct promoters, but which are identical in their coding regions. These transcripts are differentially expressed through development. Putative binding sites for hypoxia-regulated transcription factors have been identified in the promoter regions. In fact, the *Drosophila* hemoglobin is down-regulated in response to hypoxia. The exact function of this protein remains to be established, but it is conceivable that the hemoglobin serve as an oxygen-buffer that prevents the cells from oxidative stress.

Effects of some antibiotics on the total protein content of the endoparasitoid *P. turionellae* L. (Hymenoptera: Ichneumonidae) pupae

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Effects of gyrase inhibitors novobiocin, nalidixic and oxolinic acid on the pupal wet weight and total protein content of hymenopterous endoparasitoid, *Pimpla turionellae* L. were investigated by rearing the larvae aseptically on chemically defined synthetic diets. Higher dietary levels of the antibiotics tested had positive effects on the total protein content of this parasitic wasp. Oxolinic acid, at all levels tested, had the most positive effects on the protein synthesis of the young pupae. A dietary level of this antibiotic, at 2.25 mg, caused an increase in the total protein content by about 30 per cent. However, this antibiotic did not affect the wet weight of these pupae. The highest level of novobiocin also increased the protein content of the pupae, whereas its lower levels had no significant effects. Inversely, the low levels of novobiocin significantly increased wet weight of the pupae but had no effects on the protein content. The highest tested level of nalidixic acid caused a significant decrease in the wet weight while it increased total protein content of the pupae.

Infestins, blood coagulation inhibitors from insect *Triatoma infestans*, are always the answer in the blood feeding process

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Haematophagous animals are a rich source of anti-hemostatic and anti-inflammatory substances, since they have to deal with their host´s inflammatory and hemostatic systems. Recently, we have cloned several Kazal-type serine protease inhibitors from midgut of *Triatoma infestans* bug, which are encoded by one gene composed by seven Kazal-type domains in tandem. In this work, we describe the purification and characterization of recombinant infestin 4, a serine protease inhibitor with a single Kazal-type domain. Infestin 4 DNA sequence was derivated from infestin gene and its molecular mass is about 6 kDa. Infestin 4 inhibited factor XIIa (Ki = 0.1 nM), factor Xa (Ki = 53 nM), plasmin (Ki = 2.1 nM) and trypsin (Ki = 11 nM). Infestin 4 prolonged the TTPA coagulation time in about 3 times by inhibiting factor XIIa. We also identified and partially purified a native factor XIIa inhibitor from the anterior midgut of *T. infestans*. We believe that infestins perform a role in the *T. infestans* midgut during meal acquisition and digestion mainly because different infestins (1-2 and 4) present in the midgut can control blood coagulation inhibiting thrombin, factor Xa, and factor XIIa activities. Supported by FAPESP and CNPq.

Antioxidant and detoxifying enzyme activities in two strains of *Oncopeltus fasciatus* (Heteroptera: Lygaeidae) exposed to cadmium: II. chronic exposure.

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Catalase (CAT), general esterase (EST), glutathione reductase (GR), glutathione S-transferase (GST), peroxidase activity of glutathione S-transferase (GSTpx), and superoxide dismutase activity (SOD) were determined in adult *O. fasciatus* exposed to Cd in inking water from egg eclosion to adult moult. Two different strains were studied: OS, a laboratory-reared strain, and OC, a strain subjected to Cd selection, form which two consecutive generations, F_{49} and F_{50}, were analysed. Differences in median lethal mortalities (LC_{50}, defined as failure to reach the adult stage) were observed between strains, and also between early and late insects within each OC generation; recorded LC_{50}s were 25 mg Cd/l for OS, 83/36 mg Cd/l for early/late F_{49} insects, and 62/52 mg Cd/l for early/late F_{50} insects, respectively. All enzyme activities were slightly increased in F_{49} and early F_{50} insects when compared to OS insects, whereas they were slightly decreased in late F_{50} insects. Chronic exposure to Cd caused an increase in CAT and GST activities in all strains. EST activity was also increased in OS and F_{50} exposed insects. No clear pattern was observed for GR, GSTpx and SOD activities.

Dosage effects between dietary niacin and other B vitamins on larval development of *Ceratitis capitata* (Diptera: Tephritidae)

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The nutritional interaction between niacin and a nine other B vitamins group in the larval diet was evaluated. Four doses of niacin and a mixture of nine other B vitamins were cross-tested. Diets with doses ≥ 70.7 ppm of the other B vitamins and ≤0.2 ppm of niacin resulted in no pupal recovery. A delayed larval duration and reduced pupal recovery resulted when no niacin and ≤7.07 ppm of the other B vitamins in a diet. With omission of the other B vitamins, larval duration was delayed, but was not dose dependent on niacin and pupal recovery was not affected if niacin levels were maintained at ≥ 0.2 ppm. With 20 ppm of niacin in the diet, the larval duration and pupal recovery were dependent on the dose of the other B vitamins. The more the other B vitamins was, the shorter the larval durations and the more the pupal recoveries were. The other B vitamins reduced the larval duration when niacin was maintained at ≥ 2 ppm. When the niacin levels decreased to ≤ 0.2 ppm, doses of the other vitamins has to be reduced to below 7.07 ppm in order to maintain normal larval development.

Circadian rhythm of a-amylase isoform expression in *Drosophila melanogaster*

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Little is known about peripheral oscillator output in digestion. The goal of this project was to describe the circadian regulation of the a-amylase isozymes Amy-p and Amy-d in Canton-S wild type *Drosophila melanogaster*. To correct for the effect of dietary sugars which stimulate a-amylase expression, a restricted diet variant was also employed. Under control dietary conditions, in LD 12:12, amylase expression exhibited a bimodal oscillation, with a morning and early night peak. This rhythm was less robust in females than in males. In constant darkness, DD, both sexes also showed a bimodal rhythm, phase-delayed with respect to LD. Under constant light, LL, both males and females expressed a trimodal rhyhm, with an early day peak, and two smaller peaks at night. This rhythm was significant in the females but not in the males. In all cases Amy-d was expressed at a much higher level than Amy-p. Under the restricted diet, in LD, DD and LL the rhythms of expression of amylase were very similar to the control. In all cases, both isoforms were expressed at an equal level, or with Amy-p dominating, confirming the hypothesis that Amy-d expression is more strongly inhibited by glucose.

Effect of serotonin analogues on foregut contraction in the black field cricket

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Serotonin (5-hydroxytryptamine) stimulates contractions in the isolated foregut of the black field cricket, *Teleogryllus commodus* (Walker). Although several types of serotonergic receptors are known in vertebrates, whether such a diverse range of receptors is present in insects is not clear. Using analogues of serotonin characterised by their binding to specific vertebrate receptors (types 5-HT$_2$, 5-HT$_3$ and 5-HT$_4$), we investigated which receptors might be present on the foregut or associated nerve system. The foregut increased contraction to each of these analogues, although the analogue that binds to the 5-HT$_2$ receptor appeared to have the greatest effect. As immunohistochemistry indicates that the ingluvial ganglion and nerves going to the crop contain serotonin, we suggest that this might be the normal chemical released following feeding that initiates crop movement and food mixing.

A pH-dependent shift in protein digestion in *Tenebrio molitor* larvae

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Anterior (AM) and posterior (PM) parts of *Tenebrio molitor* larval midgut differ significantly in pH: 5.6 and 7.9, respectively. This difference is reflected in the proteinase sets in AM and PM. Cysteine proteinases (CP) presented by 4 fractions are dominant in AM. All fractions display maximal activity at pH 5.0-7.0, are stable at pH 5.6 and loose up to 80% of activity at pH 7.9. Their instability in PM-like environment makes very likely participation of CP in performing and regulation of only initial stages of food proteolysis. The major part of PM proteolytic activity belongs to serine proteinases. The most active trypsin-like proteinase is a cationic protein located predominantly in PM. Three anionic trypsin-like activities are distributed more evenly between both midgut parts. The main chymotrypsin-like proteinases are presented by 1 cationic and 4 anionic active fractions. Two of them including the cationic one are located mainly in PM while others are evenly distributed in AM and PM. pH-optima of serine proteinases are between 7.0 and 10.0. Thus, digestive proteolysis in *T. molitor* larvae depends on pH and is performed by a wide range of different proteinases. The work was supported by RFBR (02-04-48808) and CRDF (RB2-2396-MO-02).

Bemisia tabaci B biotype whitefly cathepsin B-like proteases

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Bemisia tabaci B biotype whiteflies are harmful to a variety of crop species because, as vectors of plant viruses, they ingest virions as they feed on the phloem of infected plants, and transmit them when they feed on uninfected plants. As part of our ongoing interest in the digestive systems of plant feeding insects, we have demonstrated significant activity of cysteine proteases at the protein level. However, our most recent findings suggest that there is a mixture of more than one family of proteases. Because we know that the proteolytic pH optimum in whole whitefly extracts is at pH 5.5, we feel that the proteases must be able to function in acidic environments. Using Cathepsin B, an acidic cysteine protease commonly found in hemipterans, as a positive control, we have optimized our protease inhibition assays. Based on the knowledge of a cDNA for the same enzyme from a related homopteran rice pest, *Nilaparvata lugens*, and related insect sequences, we have also designed PCR primers that have allowed us to 'fish' in genomic DNA preparations. We hope that future experiments will yield products that will allow us to develop probes that will eventually lead to a characterization of the gene.

Changes in haemagglutination activity in the different developmental stages of *Culex pipiens* s.l. caused by insect hormones

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Effects of methoprene and 20-hydroxyecdysone on development and haemagglutination activity (HA) were studied in both sexes of anautogenous *C. p. quinquefasciatus* and autogenous *C. p. molestus*. Methoprene and 20-hydroxyecdysone induced larval mortality, prolongation of the intermolt period in preimaginal stages, morphological changes in metamorphosis and changes in HA. HA was found in larval and adult stage of both sexes. Our results show that HA in the adult female was not dependent on blood sucking, but on the age of the females. This conclusion has been confirmed by the results obtained with autogenous *C. p. molestus,* where HA does not change in hungry or engorged females. The conclusion has been further confirmed by findings of high HA in the gut of last larval instars, developmental stages that never take blood. Methoprene caused a significant decrease of HA in the gut of the last larval instar and in adults of both sexes. On the other hand, 20-hydroxyecdysone decreased HA only in the female gut. Results obtained indicate that HA depends on the sex, on the studied organ and on the level of hormones and could be induced in different ways, not only by ingesting of blood or protein meal.

Transferrin in *Galleria mellonella*: molecular characterization and regulation by PAMPs

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Complete mRNA sequence of *Galleria mellonella* transferrin was obtained and compared with those of other species. Until now, two types of insect transferrins have been reported. Transferrins in cockroach and termite have two iron-binding sites while those in other most insect groups, studied for the protein, have only one. It was suggested that the presence of two types of the protein was related with transferrin evolution because vertebrate transferrins have two iron-binding sites, called N- and C-terminal lobe respectively. From the cDNA sequence, it was shown that *G. mellonella* transferrin also has only one iron-binding site (N-terminal lobe), and the sequence of the protein was most similar to those of *Manduca sexta* and *Bombyx mori*. In addition, regulation of transferrin mRNA and the protein itself was detected by semi-quantitative RT PCR and Western blot analysis after injection of pathogen-associated molecular patterns (PAMPs) into larval hemolymph.

Immune-induced tyrosine 3-hydroxylase gene in eri-silkworm, *Samia cynthia ricini*

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We have studied the genes that are differentially expressed in the fat body of *Samia cynthia ricini* after bacterial challenge using suppression subtractive hybridization (SSH) method. One of the cDNA fragments from the subtracted cDNA library had high homology with tyrosine 3-hydroxylase (TH), and the full length cDNA was obtained by 3' and 5'RACE methods. TH is the rate-limiting enzyme in the synthesis of catecholamine, which catalyzes the oxidization of L-tyrosine to L-dopa in the presence of tetrahydropteridine as cofactor. TH has not known so far to be involved in any of immune responses in insect, and is apparently different enzyme from phenoloxydase which similarly catalyzes the oxidation of L-tyrosine to dopaquinone via L-dopa. The deduced amino acid sequence of *Samia* TH had approx. 75% and 50% homology with those of *Drosophila* and rat TH, respectively. The *Samia* TH gene was induced in the fat body, midgut and hemocyte 9 to 18 hr after injection of bacteria.

Suitability of five danaid butterflies for parasitization by the tachinid, *Sturmia bella*

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The tachinid fly, *Sturmia bella*, is a principal parasitoid of the migrant danaid, *Parantica sita* in Japan. Female adults deposit microtype eggs on foliage of host's food plants, and parasitization starts by host ingestion of eggs with leaf tissue. Eggs of *S. bella* were found on leaves of *Hoya carnosa, Tylophora tanakae, Parsonsia laevigata, Cynanchum liukiuense* and *Ficus microcarpa,* host plants of 5 danaids, *P. sita, Ideopsis similis, Idea leuconoe, Salatura genutia* and *Euploea mulciber,* respectively, in Ryukyu Islands. However, there have been few records of parasitization of these danaids except *P. sita*. In this study, we gave a piece of artificial diet with *S. bella* eggs to 4th or 5th instar larvae of the 5 danaids to compare parasitism rates. Most of *P. sita* and *E. mulciber* larvae died from parasitization, while most individuals of *I. similis, I. leuconoe* and *S. genutia* emerged as healthy-looking adults. We found melanized larvae of *S. bella* within the abdomen of most *I. similis* adults emerging, while no sign of parasitization was observed inside the body of *I. leuconoe* and *S. genutia.* The results show that larvae of *I. similis, I. leuconoe* and *S. genutia* have the ability to overcome the parasitization by *S. bella* and develop into adults. We will discuss the relationship between migratory activities and food plant preference of these danaids from a view of defence ability against parasitoids.

Clip-domain serine proteases involved in *Manduca* immune reactions

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Clip-domain serine proteases are involved in insect immune reactions such as the Toll pathway (induction of antimicrobial peptides) and the phenoloxidase activation cascade. We identified clip-domain serine proteases HP6, 8, 15, 17 and 21 from the tobacco hornworm, *Manduca sexta* that are cleaved after stimulation with microbial elicitors. HP6 is similar to *Drosophila* persephone, which is involved in the Toll pathway in antifungal response. HP8 is similar to *Drosophila* easter, which acts in the Toll pathway by cleaving prospätzle in dorsal-ventral patterning. HP15 and HP21 are similar to *Manduca* prophenoloxidase activating protease (PAP) 2. HP17 is similar to *Drosophila* CG1299, whose function is not known. We analyzed functions of HP6 using an inactive mutant protease, in which the active site Ser was substituted with Ala. The mutant protease inhibited cleavage of HP8 stimulated by elicitors *in vitro* and suppressed induction of antimicrobial peptides *in vivo*. The results suggested that HP6 is located up-stream of HP8 in an immune reaction cascade that stimulates antimicrobial peptide synthesis. Immunoprecipitation of plasma samples with anti-HP17 antibody inhibited activation of HP21 *in vitro*, suggesting HP17 is upstream of HP21 in an immune reaction cascade.

"Recent climate [AGM2]warming promotes the northward invasion in the swallowtail butterfly, *Papilio memnon* in Japan"

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The swallowtail, *Papilio memnon*, is distributed widely from tropical to temperate zones in the Oriental region, and the northern limit of its distribution lies in western Japan. Recently the distribution has been expanding to northeastern areas in central Honshu. To investigate the relationship between the phenomena and characteristics of pupal diapause of this species, the photoperiodic response of 4 Japanese populations including the subtropical and northernmost ones was examined. Also we compared cold hardiness of pupae among the 4 populations. The critical photoperiod for inducing pupal diapause at 20C was 12 h 30 min in the subtropical population, and about 13 h in the 3 temperate ones. However, the differences in diapause intensity (about 100 days) and supercooling point (about -21C) among the 4 populations was small. Thus it may be difficult to explain the change in the distribution of this species from the change in characteristics of pupal diapause and cold hardiness of pupae. These results show that rising winter temperatures may be responsible for the recent northward invasion of this species.

The fate of foreign proteins in tobacco budworm, *Heliothis virescens***.**

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The oral delivery of protein insecticides to target sites in the insect hemocoel is made difficult due to degradation in the gut by endo- and exo-peptidases and their limited movement across the gut epithelium. Overcoming these obstacles to using protein toxins as insecticides requires better understanding the physiology of protein movement and fate in pest species. In previous studies we used Bovine Serum Albumin (BSA) and anti-BSA polyclonal antibody as model polypeptides to determine protein fate in tobacco budworms, *Heliothis virescens.* Oral delivery of anti-BSA or BSA results in the appearance of both proteins in the hemolymph over time. In contrast if anti-BSA or BSA are injected into the hemocoel both proteins turnover with time through protease degradation and/or tissue sequestration. The accumulation of foreign proteins in insect hemolymph reflects the competing rates of transfer from the diet across the midgut and turnover in the hemolymph. These results were extended to include loss of anti-BSA and BSA over time in the insect whole body. This identifies any protein sequestration that might have reduced the protein concentration in the hemolymph. Finally individual tissues were dissected after injections and tested for the presence of both proteins to determine where proteins are sequestered.

Stage-dependent expression of *Chelonus inanitus* polydnavirus genes in the host & the parasitoid

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Successful parasitism of many endoparasitic wasps depends on the supporting action of symbiontic polydnaviruses. These unique viruses have a segmented genome and are produced in the ovary; at parasitization they are injected along with the parasitoid egg into the host. In the parasitized host polydnaviruses do not replicate. In its proviral form the polydnavirus is integrated into the wasp's genome. *Chelonus inanitus* induces in its host a precocious onset of metamorphosis and a developmental arrest in the prepupal stage. The polydnavirus prevents encapsulation of the parasitoid and causes the developmental arrest. We report that the expression of six genes located on five fully sequenced segments is stage specific. Two genes are expressed only in the early phase of parasitization, two in both the early and late stages, one in the middle phase and one only at the end of parasitization. This suggests that expression of viral genes is regulated by factors from the host. For four genes the tissue distribution of transcripts was measured and it was found that the majority is located in haemocytes and fat body. Some viral genes are also expressed, although at low levels, in the parasitoid larva and pupa, i.e. from the proviral form.

Immune activation of apolipophorin-III and its distribution in hemocyte from *Hyphantria cunea*

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Apolipophorin-III is a hemolymph protein whose function is to facilitate lipid transport in an aqueous medium. Recently, apolipophorin-My Presentation for in *Galleria mellonella* larvae was shown to play an unexpected role in insect immune activation. We identified the cDNA sequence of *Hyphantria cunea* apolipophorin-III by oligonucleotide-primed amplification, and 5′- and 3′-RACE PCR. Since *H. cunea* has an unusually low level of apolipophorin- III in the hemolymph, a recombinant apolipophorin- III was overexpressed using a baculovirus expression system to investigate its biological activity. Recombinant apolipophorin- III and *E. coli* were injected into the hemocoel of last instar larvae, and the expression of antimicrobial peptide from fat body was determined by Northern blot. Injection of apolipophorin- III as well as *E. coli* induced slight up-regulation of its transcription rate in fat body. While the expression of antimicrobial peptide amatically induced by the injection of apolipophorin- III and *E. coli*. *H. cunea* hemocytes had apolipophorin- III in the granules and expressed its transcript, albeit at a much lower level than in the fat body. Upon bacterial injection, a subpopulation of hemocytes showed degranulation and degradation. Local discharge of apolipophorin- III from hemocytes caused by the injection of *E. coli* should be related to the immune response through an unknown mechanism.

Development of innovative anti-infective drugs derived from insects

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Insects are major players of terrestrial ecosystems forming the largest and the most cosmopolitan group of metazoans. A key factor that has contributed to this success is the extraordinary chemical versatility of these animals. Insects are amazingly adapted to a multiplicity of ecological niches that they colonize. Insect have the capacity to manufacture a broad range of biologically active molecules, including peptides and small molecules, which ultimately may find applications in a wide range of therapeutic fields. They can survive in inclement infectious environments which reflects their capacity to mount a potent immune response and to produce defence molecules with strong antimicrobial properties. These natural substances, either peptides or small molecules, represent promising templates to derive innovative classes of therapeutics to face the declining efficacy of classical antibiotics.

Molecular cloning and expression of aquaporin from the silk gland of *Bombyx mori*

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We have explored the system regulating pH and water in the silk gland, because this gel-like solution is maintained within the glandular lumen without irreversible coagulation and/or denaturation until the completion of spinning. In this study, to elucidate the water regulation in the silk gland, we have cloned the cDNA encoding the putative aquaporin (AQP) with a homology cloning strategy. Using degenerate primers with cDNA of anterior silk gland (ASG) followed by 5′- and 3′-RACE PCR techniques, we have obtained one candidate with homologous to other insect AQPs. The deduced amino acid sequence of AQP from ASG showed 50-60% identity with several insect AQPs such as that from *Aedes aegypti*, *Haematobia irritans exigua* and *Cicadella viridis*. The AQP expression in the ASG correlates well with the growth and development of the silk gland during the final larval instar, reaching to maximum level at the start of spinning, then declined quickly. This homologus transcript (~2.3 kb) of putative AQP from ASG was distributed not only in the midgut and the rectum, but also less in Malpighian tubules and rather lower in the tracheal systems.

Functional structure of the silk apparatus in the comb-footed spider, *Achaearanea tepodariorum* (Araneae: Theridiidae)

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It has been known that functional specialization of silk producing apparatus involving precise modifications of the shape of the spinnerets, the number and morphology of spinning tubes, and anatomical characteristics of the silk glands. Although the spinning apparatus often undergoes adaptative variations, some basic familial characteristics usually remain unchanged, so their shape and number of spinning apparatus can also classify spiders. Here we describe the fine structural organization of the silk glands and its spinning apparatus in the comb-footed spider, *Achaearanea tepodariorum*, with light and electron microscopes. The silk glands of the adult female spider were located in seven groups on the spinnerets including each pair of major and minor ampullates, tubuliforms, flagelliforms, aggregates, pyriforms and aciniforms. Each group of silk gland feed silk into one of the three pairs of spinnerets. Commonly the secretory silk is synthesized from rough endoplasmic reticulum (rER) of glandular epithelial cells and is transported from the rER into the secretory vesicles which are grown up by fusion with the surrounding small vesicles including the secretory silk.

The entry of polyembryonic egg-larval parasitoid into host embryo *in vitro*

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Copidosoma floridanum is a polyembryonic egg-larval parasitoid of the plussine Lepidoptera. The *C. floridanum* egg cleaves holoblastically to form a morula that is spherical mass of embryonic cells surrounded by a polynucleate membrane. The embryo proliferates clonally in the growing host larva. In this species, the female oviposits her eggs in host eggs of any stage of development. When 30-36 hour–old host eggs were parasitized, 75 % of *C. floridanum* eggs were laid in vitellophages of host *Thysanoplusia intermixta* eggs (outside of the host embryos), although the final percentage parasitism at adult emergence was 83%. To determine whether or not *C. floridanum* embryos secondarily enter into the host embryo, we examined *C. floridanum* embryos co-cultured with host embryos. Histological observations showed that the morula-stage embryo of *C. floridanum* had mobility and could enter the host embryo. The entry of foreign objects into *T. intermixta* embryos was not observed with latex beads or other parasitoid *Ascogaster ridiculatus* eggs *in vitro*, suggesting that *C. floridanum* embryos actively enter the host embryos. This study also suggested that the embryonic entry into the host embryos was accomplished by different ways according to the age of the host eggs when parasitized.

Immunization of *Manduca sexta* larvae with *E. coli* causes immunity to subsequent infection by *Photorhabdus luminescens*: induction of antimicrobial activity in haemolymph and the cost of defence.

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Injection of living cells of the normally non-pathogenic Gram negative bacterium *E. coli* into fifth instar larvae of the lepidopteran *Manduca sexta* causes subsequent strong immunity to challenge by the normally extremely pathogenic Gram negative bacterium *Photorhabdus luminescens*. Immunity is evident after only one hour and is fully developed by 12 h. We show that immunization with *E. coli* induces increased expression of a factor in haemolymph that prevents the growth of *Photorhabdus* in vitro. This is at least in part due to the presence of antimicrobial peptides. Immunity is not due to lysozyme, although this enzyme is expressed at elevated levels following immunization. The results of RNAi knockdown experiments on these and other known insect immune-related genes will be described. These results indicate that the many virulence genes present in the *Photorhabdus* genome are probably not redundant, but are required in an ecological context where host insects are not bacteriologically naïve, as in the laboratory situation. They also show that *Manduca* normally does not express its many defences against microbial infection, presumably because to do so would be costly. We describe attempts to estimate the cost of such antimicrobial defences.

In vitro and *in vivo* activity of synthetic antimicrobial peptides designed based on the insect defensin

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Synthetic antimicrobial 9-mer peptides were designed from the amino acid sequence of an active site of insect defensin to increase the number of positively charged amino acid residues. These peptides, RLRLRIGRR-NH_2, RLLLRIGRR-NH_2, RLYLRIGRR-NH_2 and ALYLAIRRR-NH_2, showed strong antimicrobial activity against bacteria and fungus. The peptides showed no growth inhibition activity against murine fibroblasts or macrophages and no hemolytic activity against rabbit erythrocytes *in vitro*. Furthermore, the administration of these peptides protected mice from a lethal methicillin-resistant *Staphylococcus aureus* (MRSA) challenge. In addition, the peptides suppressed tumor necrosis factor alpha (TNF-) gene expression and production induced by lipopolysaccharide (LPS) or lipoteichoic acid (LTA) in murine macrophages, providing an interesting problem of the contribution to protection of mice from lethal MRSA infections *in vivo*. The results suggest that these peptides are potential candidates for future therapeutic agents against antibiotic-resistant bacterial infection.

Isolation and characterization of serine proteases inhibitors from tick, *Boophilus microplus*

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The present work describes the isolation and characterization of several protease inhibitors from hard tick *B. microplus*: larvae, eggs and egg washed solution. These inhibitors were purified by classical purification methods. The purified inhibitors from larvae were named BmTI A to E and BmTI 1 to 6; they showed molecular masses between 7.5 and 18 kDa and inhibited trypsin, neutrophil elastase and human plasma kallikrein (HuPK). The best inhibitors for neutrophil elastase (Ki = 12 nM) and HuPK (Ki = 0.5 nM) were BmTI 3 and BmTI D, respectively. The N-terminal sequences of BmTI D, BmTI 2, and BmTI 3 allowed us classify them in Kunitz-BPTI inhibitor family. Two subtilisin and neutrophil elastase inhibitors (BmSIs) from eggs were purified, they presented molecular masses around 7 kDa and dissociation constants (Ki) in nM range for both enzymes. Finally, neutrophil elastase and trypsin inhibitory activities were also observed in egg washed solution. Serine proteases secreted by *Metarhizium anisopliae* fungal in culture medium were inhibited for BmTIs and BmSIs. These results suggest that BmTIs and BmSIs can have a role in the tick defense mechanisms against fungal infection. Supported by: FAPESP and CNPq.

Change of ferritin of wax moth, ***Galleria mellonella*** **on dietary heavy metal ions**

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A tissue ferritin (T-ferritin) was purified from the haemolymph-free whole body extract of wax moth, *Galleria mellonella*. This protein was distinguished from haemolymph ferritin (H-ferritin) in that it had an additional subunit of 34 kDa. H-Ferritin of this moth has been known to be composed of three subunits of 26, 30, and 32 kDa. However, the sequence of N-terminal 20 amino acid residues of the tissue specific 34 kDa polypeptide was shown to be identical with that of 32-kDa subunit of *G. mellonella* H-ferritin. It was also observed that the T-ferritin had cross-reactivity against anti-haemolymph ferritin serum. T-Ferritin was detected in fat body, gut, Malpighian tubule, and haemocyte by Western blot analysis. T-Ferritin responded differently from H-ferritin to the heavy metal injection. T-Ferritin was induced earlier than H-ferritin by $HgCl_2$ injection. T-Ferritin was sensitively increased by Cd^{2+}, Cu^{2+}, Mn^{2+}, Zn^{2+}, and Ni^{2+} ions. Hg^{2+} was a highly effective ion for the H-ferritin induction. These results suggest that there are different response mechanisms to heavy metals between T-ferritin and H-ferritin.

Proteome analysis of brush border membrane vesicles from the lightbrown apple moth, *Epiphyas postvittana*

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Brush border membrane vesicles (BBMV), formed from intestinal villae, were prepared from the midgut of fifth instar lightbrown apple moth (*Epiphyas postvittana*) larvae by differential centrifugation in the presence of a protease inhibitor cocktail. These were lysed using water, and the contents collected to give a first extract. The BBMV membranes were then sequentially extracted with a standard urea/CHAPS buffer then with thiourea/urea/SB3-10 buffer to yield two further protein extracts of membrane proteins. The extracts were separated by two-dimension gel electrophoresis (2-DE) on 7cm NuPage Novex 4-12% Bis-Tris Zoom gels using pH ranges of 4-7 and 3-10. For protein identification, protein spots on gels stained with colloidal Coomassie Brilliant Blue G-250 were excised, digested in gel with trypsin, and analysed by peptide mass fingerprinting after electrospray MS-MS mass spectrometry. The 2-DE of Extract 2 was heavily degraded, with few protein spots greater than 20 kDa, suggesting that there was trouble with proteases despite the addition of a protease inhibitor cocktail. Extracts 1 and 3 contained many distinct spots. A number of these have been identified and include ATPase components, proteases, glycosidases, ribosomal and heat shock proteins.

Hsp70 and Fib-L are over-expressed proteins in immunized *Bombyx mori* larvae

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Hemocytes play a major role in insect cellular immune response. In infected with foreign invaders, hematopoiesis was more actively occured. The current study was performed to understand hemocyte differentiation patterns and expressed proteins of immunized *Bombyx mori* larvae. Heat-inactivated bacteria (*Bacillus megaterium*) was injected into *B. mori* 5th instar 4 day after ecdysis. After six hours, circulating hemocytes were examined. The THC were increased of 905×10^3 in immune response. The DHC indicated that granulocytes were increased of 687.5×10^3 and accounted for half of THC in immunized larvae. These results indicated that hematopoiesis were occurred actively and granulocytes were increased prominently in immunized *B. mori* larvae. Hemocytes and hemolymph proteins observation revealed that some proteins were newly expressed or up-regulated in immune response. In hemocytes, over 300 spots were counted, only one spot was newly detected and four spots were up-regulated in immunized larvae. In hemolymph proteins, about 200 spots were detected and three spots were appeared newly. Heat shock 70kDa protein (hsp70) was one of the hemocyte up-regulated proteins and fibroin light chain precursor (Fib-L) was among the hemolymph new entry proteins. Hsp70 and Fib-L were purified from immunized larvae and carry out an experiment on larvae with these proteins. These results suggest that hsp70 of hemocytes and Fib-L of hemolymph are associated with the regulation of THC and GR increasing in immune response.

Utilisation of protein in litter dwelling larvae *Bibio marci* **(Diptera: Bibionidae)**

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Saprophagous invertebrates may play crucial role in modification many ecosystem processes by its influence on decomposition and soil structure. Leaf litter contains hardly utilisable substances and is deficient for protein. Moreover protein is typically immobilised by phenols of humic-acids. Protease pH optima, its distribution in the gut, distribution of amino-acids and ammonia in gut compartments were studied in bibionid larva *Bibio marci*. Protein digestion was studied using ^{14}C-labelled protein either pure protein or as protein stabilised by humic-acids were used. Contribution of larvae to protein mineralisation was estimated as proportion of protein mineralised in the presence of larvae and in control litter. Protease showed alkaline pH optimum corresponding with pH in anterior gut. Trypsin activity was high in caeca, anterior and mesenteron but lowered in hindgut. Content of amino-acids decreased inside gut, ammonia content increased. Serine and valine proportions decreased, methionine proportion increased during gut passage. High methionine proportion occurred in hindgut. Larvae utilised both pure and humic-acid stabilised protein. Larvae used pure protein more efficiently than bounded one. Contribution of larvae to protein mineralisation is higher in bounded protein. Gut alkaline pH gut probably supported humic-acid bounded protein utilisation by larvae similarly as already documented in termites and some scarabeid larvae.

Boophilus microplus trypsin inhibitor-A (Bmti-A): is a serine or cysteine proteinase inhibitor?

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Ticks are arthropod blood feeders and to control their host's haemostatic system, they secreted coagulation and platelet aggregation inhibitors in the saliva during feeding process. Among them tick anticoagulant protein (Tap), A factor Xa inhibitor, And ornithodorin, both purified from *Ornithodoros moubata*; savignin A thrombin inhibitor and savignygrin, a platelet aggregation inhibitor from *O. Savignyi* species, all these proteins have in common the Kunitz-Bpti structure fold. Beside these anti-haemostatic molecules, several proteinase inhibitors with unknown physiological function were described in *B. microplus* and *Riphicephalus Sanguineus* species. In the present work, we describe a new function for the Bmti-A, as a cysteine proteinase inhibitor. Bmti-A belongs to the Bpti-Kunitz type family; it inhibited trypsin, chymotrypsin, plasma kallikrein and neutrophil elastase and presented molecular mass around 18 Kda, containing two kunitz domains. Our present results showed that Bmti-A strongly inhibited cathepsin L (Ki = 23 Pm), a cysteine proteinase. Confirming this data, Bpti, a kunitz type inhibitor, also inhibited the following cysteine proteinases: cathepsin L (Ki = 50 Pm), papain (Ki = 3.4 Nm) and Vtdce (0.12 Nm), a cysteine proteinase from *B. microplus* eggs. Our recently findings showed a bifunctional activity of Bpti-Kunitz type inhibitors. Supported by: Fapesp and Cnpq.

Functional analysis of BmRel proteins

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Antibacterial peptide genes are known to be up-regulated by bacterial infection in insect. In *Bombyx mori*, the B-like motifs, suggesting an important role in gene activation, are located on the 5′-upstream region of these genes. Two cDNAs encoding BmRelA and BmRelB were cloned as candidates for binding factors on B-like motifs. From the nucleotide sequence information, it is suggested BmRelA and B have the same amino acid sequences except BmRelA has extra 52 amino acids in the N terminal region. Transient luciferase assay indicated BmRelA and BmRelB differentially activated *Bombyx mori* antibacterial peptide genes. In order to identify the transcriptional activation domain of BmRel, luciferase assay experiments were carried out using deletion constructs of BmRel. Results showed proline rich region (86 amino acids) was involved in the activation of attacin gene by BmRelB. On the other hand, for the activation of lebocin4 gene by BmRelA, both proline rich region and A specific 52 amino acid were necessary. This 52 amino acid sequence also had a function as the transcriptional activation domain in yeast. This sequence was occupied by mainly hydrophobic and acidic amino acids, one of the typical amino acid sequence as transcriptional activation domain.

Alteration of cellulolytic systems in termites

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Cellulolytic systems in the gut of termites was examined by measuring hydrolysis of crystalline and carboxymethyl cellulose as well as performing reverse transcription-PCR and phylogenetic analyses of endogenous endo- -1,4-glucanase cDNAs from termites of diverse feeding habits. In cellulolytic flagellate-harbouring termites, carboxymethyl cellulose, which is a highly soluble cellulose ether, was primarily hydrolysed in the salivary glands, whereas microcrystalline cellulose was degraded primarily in the hindgut. In non flagellate-harbouring termites, a fungus-growing termite retains both the cellulolytic activities in the salivary glands, while other wood-feeding and soil-feeding termites possess both the cellulolytic activities primarily in the midgut. RT-PCR and phylogenetic analyses revealed that multiple endogenous endo- -1,4-glucanase genes are expressed in the salivary glands of the flagellate-harbouring termites as well as the fungus-growing termite and are expressed in the midgut of the wood- and soil-feeding non flagellate-harbouring termites, which are likely to be related to diversification of feeding habits in termites of the family Termitidae. In the light of these results, as well as recently accrued phylogenetic data, we discuss scenarios for the evolution of cellulose digestion in termites.

The effects of bacteria and TLCK on hemocyte morphology of *Manduca sexta*

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Conflict and war are everywhere. The microscopic world is no exception. Battle is just as necessary for survival. For *Manduca sexta*, the tobacco hornworm, invaders include parasites, bacteria, or other foreign material. Our general question is: how do *M. sexta* hemocytes (mainly plasmatocytes and granular cells) respond to threats. Hemocytes phagocytose or form nodules around bacteria; they form capsules around larger pathogens like parasites. In vitro, we looked at hemocytes from immune activated (bacteria injected) and control larvae. For immune activated compared to control larvae: plasmatocytes had significantly greater mean length; granular cells had greater mean area. At 20 hr, hemocytes from immune activated larvae organized into a uniform network substantially faster than the control cells. We also studied the effect of TLCK, a serine protease inhibitor. With immune activation, at 20 hr, TLCK resulted in: (1) greater granular cell mean area; (2) more and larger vesicles in granular cells; (3) fewer plasmatocytes connect the network nodes. Regardless of immune activation, TLCK showed no effect on plasmatocyte length at 1 hr; nevertheless, TLCK inhibited network formation at 20 hr.

"Carotenoid binding protein is encoded by Y gene which transport carotenoid into hemolymph of *Bombyx mori*.[AGM3]"

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Fundamental aspects of carotenoid absorption are largely unknown, such as luminal and intracellular factors related to absorb and transport of carotenoid. These mutants offer unique research tools for studying lipid specific transporter in insects. In the +Y gene, the transport of the other lipids except carotenoid is normal, only the transfer of lutein is affected. A carotenoid binding protein (CBP) has been isolated from *Bombyx mori* larvae. Immunoblotting analysis and immunocytochemistry confirmed the presence of CBP in larvae with the dominant Y gene only. RFLP mapping analysis shows almost certainly that Y gene encode CBP. In both Y and +Y mutants, CBP gene and CBP mRNA were found by Southern and Northern hybridization. The difference of sequences of CBP cDNA from Y and +Y strains was not found. mRNA size of +Y mutant is smaller than Y . It is not clear why +Y strain can not make CBP. $+^{Y}$ might have a mutation on the regulatory region related to CBP expression.

Immune evasion mechanisms of the endoparasitoid wasp *Hyposoter didymator* (Hym.: Ichneumonidae) in four lepidopteran host species

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The relationship between immune suppression response and host range has been evaluated in four lepidopteran hosts of the ichneumonid *Hyposoter didymator.* Parasitism inhibited the growth of permissive (*Helicoverpa armigera*) and semi-permissive hosts (*Spodoptera littoralis* and *S. exigua*), whereas the non-permissive *Trichoplusia ni* was not significantly affected. Encapsulation mechanisms were only detected in semi and non-permissive hosts. The presence and the role of polydnavirus in ovaries of *H. didymator* females was evaluated by electronic transmission microscopy. Polydnavirus were always detected, but the semi-permissive and non-permissive hosts encapsulated eggs and larvae of the parasitoid. Hyperparasitism was detected in semi and non-permissive species and in the former, one or more larvae were found floating free in the hemolymph. The host range of *H. didymator* seems to be determined by whether or not the parasitoid ichnovirus is able to establish persistent infections in parasitized host larvae to provide long-term suppression of host immunity system.

Studies about the immune response of four lepidopteran species & the immune evasion mechanisms of the parasitoid wasp *Hyposoter Didymator* (Hym., Ichneumonidae).

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To evaluate the relationship between immune suppression and host range, four lepidopteran hosts were parasitized by ichneumonid parasitoid *Hyposoter didymator.* **Parasitism inhibited the growth of permissive (***Helicoverpa armigera***) and semi-permissive hosts (***Spodoptera littoralis* **and** *S. exigua***), whereas growth in non-permissive host (***Trichoplusia ni***) was not significantly affected. Encapsulation mechanisms were detected in species semi and non-permissive but not in permissive host. In the second part of this work, the presence of polidnavirus in ovaries of** *H. didymator* **females exposed to these hosts has been evaluated by electronic transmission microscopy. Polidnavirus were detected in all tested females, although some hosts (semi-permissive and non-permissive) encapsulated eggs and larvae of the parasitoid. Moreover, hyperparasitism was detected in semi and non-permissive species but one or more larvae were detected floating free in the hemolinfa in semi-permissive host. The host range of** *H. didymator* **seems to be determined by whether or not the parasitoid ichnovirus is able to establish persistent infections of parasitized larvae to provide long-term suppression of host immunity.**

Glycosyde-hydrolases in the digestive systems of lower termites

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Since the finding of the endogenous cellulase genes belonging to the glycoside-hydrolase family 9 (GH9) from the termite *Reticulitermes speratus* in 1998 (Watanabe et al., 1998), we have found expressions of -glucosidase (GH1) in the salivary glands of the lower termite *Neotermes koshunensis*, endo- -1,4-glucanases (Tokuda et al., 2002) and cellobiohydrolase homologues (both GH7) in the hindgut parabasalian symbionts (protozoa) (Nakashima et al., 2002; Watanabe et al., 2002) by purification and N-terminal amino-acid sequencing of the enzymes, and following cDNA cloning. To elucidate further details of the lower termite digestive systems, EST analysis was employed for mRNA extracted from the salivary glands of *R. speratus*. The EST clones (about 400 at the submission) analyzed preliminarily encoded endo- -1,4-glucanase (GH9, 55.8%), -amylase (GH1, 4.7%), -glucosidase (GH1, 0.5%), -1,3-glucanase (GH16, 0.5%) and a possible glycosyl-transferase (0.5%), but no cellobiohydrolase homologue encoding cDNAs have not found. The result suggested importance of protozoan cellobiohydrolase generally responsible for degradation of crystalline region of the substrate.

Morphology & function of cuticular nanostructure arrays on insect wings-an investigation using atomic force microscopy

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The characteristics of naturally occurring nano-structured arrays has been of great interest to physicists and biologists as these evolutionary refined structures provide ideal models, or templates, for man-made fabricated materials and devices. Ordered hexagonal packed array structures on cicada (e.g., *Pflatoda claripennis*) and termite (e.g., family *Rhinotermitidae*) wings have been investigated in this study. The spacings range from ca. 200 to more than 800 nm. The structures tend to have a rounded or in some cases a near-paraboloid shape at the apex and protrude some 150 to 350 nm out from the membrane surfaces. Array structures with spacings at the lower end of the range are most likely optimised to serve as an anti-reflective coating. The structures with spacings at the upper end of the range are unlikely to offer these properties in the visible spectrum, thus indicating that they confer additional, and most likely different, evolutionary advantages to those of camouflage. Atomic force microscopy has been utilised in this study for determining topography, membrane stiffness and adhesion properties at the micro and nanometre scales. The results indicate that the array structures have multiple functions including, anti-reflective properties, self-cleaning mechanisms and structural support elements.

Members of the viral ankyrin (vankyrin) gene family in the *Campoletis sonorensis* ichnovirus (*Cs*IV) exhibit tissue-specific expression in parasitized *Heliothis virescens* larvae & differential biological effects *in vitro*.

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Polydnaviruses (PDVs) occur in obligate symbiotic associations with Ichneumonid (Ichnoviruses or IVs) and Braconid (Bracoviruses or BVs) wasps. PDVs are injected during parasitization, and viral gene products alter host physiology enabling endoparasitoid development. Our data represent investigations of a gene family (*vankyrin*) that is shared by both IV and BV genomes. Vankyrin proteins possess ankyrin repeats that resemble the inhibitory domains of NF-κβ transcription factor inhibitors (Iκβs). *vankyrin* genes from the *Campoletis sonorensis* IV (*Cs*IV) genome are expressed with different tissue specificities early during parasitization in *Heliothis virescens* larvae and can be divided into two subclasses: those that target host fat body nuclei, and those that target host hemocyte nuclei. The two different subclasses of *vankyrin* genes in *Cs*IV also exhibit different biological effects when expressed from recombinant *Ac*MNPV baculoviruses *in vitro*. Insect cell lines infected with viruses expressing fat body specific genes exhibit enhanced longevity compared to those infected with viruses expressing hemocyte specific genes. Preliminary electrophoretic mobility shift assays indicate that *Cs*IV vankyrin proteins can disrupt the ability of nuclear extracts to bind insect NF-κβ target sites *in vitro*. We hypothesize that *Cs*IV *vankyrin* proteins may suppress NF-κβ activity during immune responses and developmental cascades in parasitized hosts.

Using flow cytometry to determine differential hemocyte counts in suitable and unsuitable stemborer hosts parasitized by *Cotesia flavipes*.

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We used flow cytometry to determine differential hemocyte counts in partially suitable and unsuitable hosts (*Diatraea grandiosella* and *Ostrinia nubilalis*, respectively) parasitized by novel-association *Cotesia* spp. parasitoids around the time encapsulation occurred and compared results with those from the suitable host (*Diatraea saccharalis*). Flow cytometry has been useful to rapidly classify individual cells based on light scatter and fluorescence emission characteristics. The autofluorescence emission patterns of host hemocytes were analyzed, as well as emission patterns after labeling with florescein peanut agglutin (PNA). PNA selectively labels granular cells thought to be important in the host's immune response, but does not label plasmatocytes. We determined that flow cytometry is a useful method for studying the hemocytes of different lepidopteran species. Using this method we determined that there are clear differences in the proportions of plasmatocytes and granular cells depending on host species and developmental stage. However, differences in hemocyte proportions did not correlate with successful parasitization or the occurrence of encapsulation. In the future we will try to determine if functioning of the hemocytes involved in the immune response is altered after parasitization of suitable and unsuitable hosts.

Cytotoxic activity in the colleterial glands of *Antheraea yamamai* **against rat hepatoma cells**

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Female insects, on oviposition, secrete a viscous fluid from their reproductive accessory glands, i.e., the colleterial glands, for fixing eggs. So far, some compounds have been found in the secretions of several species. For example, in praying mantises, phenolic compounds and enzymes have been found in addition to structural proteins as a main component, which are involved in the hardening of the ootheca or the eggcase (Yago *et al.*, 1984). Recently several anti-bacterial polypeptides have been found in the glandular secretions of flies (Marchini *et al.*, 1989, 1991, 1997; Tirone *et al.*, 2000), which protect the genital tract, eggs and larvae from microbial invasion. These findings suggest that such secretions in other species may also contain some new and bioactive substances. Thus, the colleterial gland of the Japanese wild silkmoth, *Antheraea yamamai,* was examined. When a crude extract of the glands was added to a suspension of rat hepatoma cells (Lh84 cell line), a majority of the cells were found to be dead after 24 h of incubation. The extract was further purified using HPLC and fractions were obtained exhibiting the same activity. The characterization of the active substance is currently under way.

Effects of venoms from two different species of pteromalid wasps on the spreading, viability & encapsulation capacity of *Pieris rapae* hemocytes

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Effects of venom from a pupal-stage endoparasitoid, *Pteromalus puparum* (Hymenoptera: Pteromalidae) on hemocytes of its host *Pieris rapae* pupae both *in vivo* and *in vitro* were measured by comparison with venom from a pupal-stage ectoparasitoid, *Nosonia vitripennis* (Hymenoptera: Pteromalidae) with no ability to parasitize *Pieris rapae* in nature. The spreading and viability as well the capability to encapsulate Sephadex A-50 beads of *Pieris rapae* pupae hemocytes were significantly reduced after treatment by *Pteromalus puparum* venom. The medium concentrations of the venom to inhibit encapsulation of host hemocytes *in vivo* and in *vitro* were 0.0425 VRE/pupa and 0.00007 VRE/µl, respectively. In contrast, *Nosonia vitripennis* venom did not appear to have any effect on *P. rapae* hemocytes either *in vivo* or *in vitro* under conditions as the same as treated by *P. puparum* venom. To compare these different findings, it is apparent that the physiological effect of the venom from a defined species against insect target hemocytes has its host specificity.

"Characterization & cDNA sequence of lysozyme of *Agrius convolvuli*"

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A new family member of insect lysozyme, an antibacterial peptide, has been isolated from the hemolymph of the fifth instar larvae of *Agrius convolvuli*. *Agrius* lysozyme was isolated by cation-exchange chromatography and RP-FPLC, and sequenced by HPLC system. It was observed that the purified *Agrius* lysozyme was heat-stable and had a molecular weight of approximately 15 kDa by SDS-PAGE. The N-terminal amino acid sequence of *Agrius* lysozyme is K-H-F-S-R-C-G-L-V-Q-E-L-F-W-Q-G-F-P with the highest similarity to that of *Heliothis virescence*, and it has been identified that the *Agrius* lysozyme belongs to *c* type lysozyme. A cDNA clone encoding a lysozyme was isolated and its complete sequence was determined. The coding region indicates that this lysozyme consists of a 19 residues signal peptide and a 120 residues mature protein which is very similar to the other lepidopteran lysozymes. Total numbers of negatively charged residues (Asp and Glu) were 13 and positively charged residues (Arg, Lys and His) were 22. Eight Cys residues were conserved in the same positions as that of other insect lysozymes.

Purification and cloning of lysozyme from hemolymph of *Spodoptera litura*

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Lysozyme is one of the components of the innate immunity of the insects. The lysozyme has been isolated by heat treatment, cation exchange, and reversed-phase chromatography from immunized hemolymph of last instar larvae of *Spodoptera litura*. Lysozyme level in immunized hemolymph was measured by ECL western blot analysis and was very high at 48 hours after injection with E. coli K12 (1×10^7). The molecular weight of *Spodoptera* lysozyme was estimated to be about 15 kDa by SDS-PAGE and it is great similarity with *Agrius* lysozyme, which recently purified from larval hemolymph of *Agrius convolvuli*. The 3' and the 5' regions of the lysozyme gene were amplified using the PCR protocol for the rapid amplification of cDNA ends (RACE). We will discuss this lysozyme.

The purification & characterization of a lipid transfer particle from wax moth, *Galleria mellonella* L.

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A lipid transfer particle (LTP) was purified from the larval hemolymph of wax moth, *Galleria mellonella* by two step KBr density gradient ultracentrifugation, anion exchange chromatography (DEAE-Trisacryl M), YM-50 ultrafiltration (Amicon) and gel permeation chromatography (Sephadex G-100). LTP is composed of three subunits (apoLTP-I, apoLTP-II, and apoLTP-III). Molecular weights of each subunit were determined (apoLTP-I = 280 kDa, apoLTP-II = 84kDa, and apoLTP-III = 55kDa). The confirmation of LTP was determined by western blotting with polyclonal antibody for *Bombyx mori* LTP because LTP is non species – specific. ApoLTP-I and apoLTP-III of *G. mellonella* react intensively with that of *B. mori* LTP whereas apoLTP-II react weakly with that of *B. mori* LTP.

Is the *Cotesia rubecula* venom protein calreticulin involved in the suppression of host defensive responses?

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Encapsulation is a major cellular defensive response of insects against intruding pathogens and parasitoids. Parasitoids have developed numerous mechanisms to suppress the host encapsulation by interfering with every step including recognition, adherence and spreading. During egg deposition, most endoparasitoid wasps inject maternal factors into hosts to suppress host immune reactions and ensure successful development of the parasitoid progeny. In previous studies, we isolated some *Cotesia rubecula* venom proteins, which inhibit melanization of host hemolymph by interfering with the pro-phenoloxidase activation cascade. Here we report the isolation and characterization of a novel venom protein, calreticulin, from *C. rubecula*. Results indicate that the protein may prevent encapsulation reactions. It is possible that the protein might function as an antagonist molecule competing for binding sites with the host calreticulin, which is involved in non-self recognition. Such inactivation might have synergistic effects in conjunction with the immunosuppressive action of a calyx protein Crp32, which protects the parasitoid eggs prior to expression of polydnaviruses.

Recent climate warming promotes the northward invasion in the swallowtail butterfly, *Papilio memnon* in Japan

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The swallowtail, *Papilio memnon*, is distributed widely from tropical to temperate zones in the Oriental region, and the northern limit of its distribution lies in western Japan. Recently the distribution has been expanding to northeastern areas in central Honshu. To investigate the relationship between the phenomena and characteristics of pupal diapause of this species, the photoperiodic response of 4 Japanese populations including the subtropical and northernmost ones was examined. Also we compared cold hardiness of pupae among the 4 populations. The critical photoperiod for inducing pupal diapause at 20C was 12 h 30 min in the subtropical population, and about 13 h in the 3 temperate ones. However, the differences in diapause intensity (about 100 days) and supercooling point (about -21C) among the 4 populations was small. Thus it may be difficult to explain the change in the distribution of this species from the change in characteristics of pupal diapause and cold hardiness of pupae. These results show that rising winter temperatures may be responsible for the recent northward invasion of this species.

The emergency life-history stage and immunosuppression in insects

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Events such as the appearance of a predator require an immediate, coordinated response from any animal, including insects. In vertebrates and mollusks, part of this rapid, emergency response typically involves a change in immune function, although the adaptive function of these changes remains unclear. In male crickets (*Gryllus texensis*) short bouts of flying (3 min, n=63), restraint stress (10 min, n=126), or cold stress (4° C for 10 min, n=69) resulted in increased susceptibility to the bacterium *Serratia marcescens* relative to controls. The immunosuppression lasted for approximately 20 min, depending on the stressor. Immediately after flight (n=12) and restraint stress (n=9), octopamine concentration increased in the hemolymph. Injections of octopamine (10^{-6} M) also reduced resistance to *S. marcescens* relative to saline injected controls. Crickets reared at high vs. low density showed no difference in their sensitivity to *S. marcescens*, nor did they differ in their baseline octopamine hemolymph concentrations. By examining how stressful events induce immunosuppression in insects, it may be possible to determine its functional significance across phyla.

Parasites may exploit a host's immune-neural connections to alter host behaviour

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The parasitoid wasp, *Cotesia congregata*, attacks the tobacco hornworm, *Manduca sexta*. Previous work has demonstrated that the inhibition of host feeding is mediated, in part, by elevated concentrations of octopamine in the host's hemolymph. The source of the excess octopamine is not the parasitoid. In this paper I show that the behavioural change in the host correlated with a decrease in the number of hemocytes in the host's hemolymph. The amount of octopamine in the cellular fraction of the hemolymph declined while the amount in the non-cellular fraction increased. Octopamine degradation in the plasma was also decreased. These results suggest that at least part of the increase in octopamine came from the host's hemocytes. Injections of lipopolysaccharides derived from *Serratia marcescens* (bacterium) increased octopamine concentrations in the hemolymph and suppressed feeding in normal *M. sexta*. Feeding was also suppressed by injecting octopamine or an octopamine agonist. An octopamine antagonist had no effect. These results suggest that the parasitoid may suppress host feeding partly by activating a possible immune-neural connection in the host.

A serine proteinase homolog venom protein inhibits activation of prophenoloxidase in insect haemolymph

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Most endoparasitic wasps inject maternal factors into their host haemocoel at parasitization to suppress the host immune system and ensure successful development of their progeny. Melanization, one of the insect defence mechanisms against intruding pathogens or parasites, is often inhibited following parasitization. We have isolated a 50 kDa protein in size from the venom of *Cotesia rubecula* (Hym: Braconidae) that blocks melanization in the haemolymph of its host, *Pieris rapae* (Lep: Pieridae). This protein, designated Vn50, is a serine proteinase homolog (SPH) containing an amino-terminal clip domain. Bacterially expressed Vn50 bound *P. rapae* haemolymph components that were recognized by antisera to prophenoloxidase (proPO) and proPO-activating proteinase (PAP). Vn50 is stable in the host hemolymph, remaining intact for at least 72 h after parasitization. Using *Manduca sexta* as a model system, we found that Vn50 efficiently down-regulated proPO activation mediated by *M. sexta* PAP-1, SPH-1, and SPH-2. Possible mechanisms of inhibition of proPO activation by Vn50 will be discussed.

Investigation on physiological effects of pyriproxyfen on greenhouse whitefly (*Trialeuroides vaporariorum* Westwood)

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Juvenoid compound pyriproxyfen is a potent suppressor of embryogenesis and adults formation of greenhouse whitefly, *T. vaporariorum.* Dipping of tobacco and tomato seedling infested with 0-1 day old eggs in 2.5 mg (AI) /liter resulted in total suppression of egg hatch. Older eggs were affected to lesser extent. Exposure of *T. vaporariorum* male and female for 48 h to tobacco and tomato leaves treated with 5 mg (A.I.)/liter resulted I oviposition of infertile eggs. This effect lesser persisted for an additional 2 day after transfer to untreated leaves. Treatment of second instars with 0.04-5 (A.I.)/ Liter pyriproxyfen resulted in normal development until suppressed in 5 m (A.I.)/Liter. At similar concentration the suppression of third instars observed. Second instars exposed to 5-25 mg (A.I)/Liter pyriproxyfen excreted honeydew at a level similar to the control until the pupation, after which reduction was observed. Inhibition of egg hatch on the lower surface of tobacco leaves was observed when their upper surface was treated with 1-25 mg (A.I.)/Liter indication a significant translational effect.

Does bioluminescence constitute an auxiliary mechanism against oxygen toxicity in click-beetles?

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A wealth of spectroscopic, chemical and biological data evidences molecular oxygen consumption to sustain the light reaction of the vast majority of bioluminescent (BL) organisms. In the case of BL beetles, all lampyrids, elaterids, and phengodids studied till now use the same luciferin to emit light, which ranges from green to red, depending on the specific luciferase tertiary structure. Interestingly, the levels of superoxide dismutase (SOD) – an important enzymatic antioxidant defence – are much lower (5x) in non-luminescent larval elaterids than in larvae of (luminescent) *Pyrophorus divergens* and respond to the pO_2 in the habitat: 3-fold higher in larval *Pyrearinus termitilluminans*, which lives in well-aerated tunnels excavated into termite mounds, than in *P. candelarius*, which dwells in decaying logs. Moreover, the bright thoracic segments of the *P. termitilluminans* larva are significantly richer (2-fold) in SOD than its abdominal dim segments. Exposure of *P. termitilluminans* larvae to hyperoxia induced both their luciferin concentration (50%) and luciferase activity (2-fold); total SOD activity also increased 50%. These and complementary data obtained from SOD, luciferase and urate studies carried out during the natural and the 20-hydroxyecdysone-induced development of larval *P. termitilluminans* suggest that bioluminescence and antioxidant systems may cooperate to minimize oxidative stress. Support: FAPESP, CNPq, PRONEX.

Host-parasitoid hormonal signaling and parasitism by *Cotesia congregata* in the tobacco hornworm *Manduca sexta*

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Hormones play a critical role in the growth and development of insect parasitoids. Host juvenile hormone and ecdysteroid levels are disrupted during parasitism of the tobacco hornworm by the gregarious braconid wasp *Cotesia congregata*. Following parasitization of host larvae in the fourth instar, the wasps emerge from developmentally arrested fifth instar larvae which have ceased feeding. Both juvenile hormone (JH) and ecdysteroids are produced by the parasitoids and secreted into the hemolymph of the host. In the host's last instar, the ecdysteroid titer of the host peaks at a level four-fold higher than the peak concentration found in prewandering unparasitized larvae, and in contrast to nonparasitized control larvae the parasitized larvae fail to expose the dorsal vessel or exhibit wandering behavior. The application of methoprene to parasitized larvae suppresses the emergence of parasitoids from the host, and also appears to interfere with the release of ecdysis-triggering hormone in the parasitoid. Results in this system will be compared to recent findings in other host-parasitoid associations.

Identification of the polydnavirus CrV1 gene homolog in *Cotesia congregata*

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The CrV1 gene of *Cotesia rubecula* polydnavirus (PDV) suppresses the cellular immune system of *Pieris rapae* larvae. Although the gene was originally described in *C. rubecula*, CrV1 gene homologs exist in several *Cotesia* species. A CrV1 homolog gene has been identified in *C. congregata* PDV. The *C. congregata* PDV not only suppresses the immune response but it may also be involved in causing hemocyte apoptosis in parasitized *Manduca sexta* larvae. Although CrV1 is expressed in a transient manner (4-8 hpp) in *P. rapae*, it is continuously expressed (from parasitization to emergence of the parasitoid wasp) in *M. sexta*. This paper reports the cloning and sequencing of the CrV1 gene homolog in *C. congregata*. The final goal is to develop a hybrid baculovirus containing the immune-suppressive gene of the PDV that can be tested against several important insect pests.

Why do insects breath discontinuously?

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The insect respiratory system consists of a branching tracheal system controlled at the openings to the atmosphere by spiracular valves. Insect physiologists have long been fascinated and puzzled by the complex control patterns observed in these spiracular valves. In particular, the Discontinuous Gas Exchange Cycles (DGC) with its Open, Flutter and Closed Phases has been the subject of much study. The most widely accepted explanation for discontinuous ventilation is that it reduces respiratory water loss. I will provide evidence in support of an additional explanation, namely that insects close their spiracles not to impede the loss of water but to reduce internal partial pressures of oxygen. The Open Phase is necessitated by the accumulation of carbon dioxide in the insect's hemolymph, tissues and tracheae. Following the Open Phase, the insect's tissues are "over-oxygenated" posing the risk of oxidative damage. The Flutter and Closed Phases serve the purpose of reducing internal oxygen partial pressures. The insect respiratory system is designed to provide oxygen and remove carbon dioxide at rapid rates during periods of high metabolic demand. At rest, DGC serves the purpose of protecting the insect's tissues from the damaging effects of elevated partial pressures of oxygen.

Oxygen transport and storage proteins in insects

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The aerobic metabolism of animals requires the supply of the internal tissues with sufficient amount of oxygen. Gas-exchange in insects and other terrestrial arthropods is mediated via trachea. For this reason, specialized O_2-transport or -storage proteins have long been regarded unnecessary in most insects, though high concentrations of hemoglobin in the millimolar range have been observed in the hemolymph of species that live under hypoxic conditions, such as the chiromonid midges, the horse botfly *Gasterophilus intestinalis*, or some aquatic Hemiptera. However, we have recently shown that intracellular hemoglobins are also present at lower concentrations (< 10 µM) in various insect species. In the fruitfly *osophila melanogaster* prominent hemoglobin expression is mainly associated with the trachea. In addition, in the hemolymph of some basal insects (e.g. Plecoptera, Thysanura), we isolated a hemocyanin that that reaches concentrations typical for arthropod hemocyanins. Oxygen binding characteristics are similar to that of crustacean hemocyanins. These observations suggest that oxygen supply in insects is more complex than previously thought, and may depend on protein-mediated transport and storage in addition to simple tracheal diffusion.

Physiological regulation of the same lepidopteran host, *Cotesia plutellae* Kurdjumov, by parasitoid's products of three different hymenopteran endoparasitoid species

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Three hymenopteran endoparasitoids, *Cotesia plutellae* (Braconidae), *Diadegma semiclausum* (Ichneumonidae) and *Oomyzus sokolowskii* (Eulophidae) were used to evaluate the effects of parasitoid's products on the lepidopteran host, *Plutella xylostella* L. *C. plutellae* injects PDVs (BVs) and venom into host during its oviposition and teratocytes are produced after parasitoid's embryos develop in host, *D. semiclausum* deliveries PDVs (IVs) and venom into host but no teratocytes are developed, while *O. sokolowskii* only has venom. The results showed that the last-instar host larvae were significantly prolonged and failed to pupate when they were parasitized by *C. plutellae*, while the hosts were also arrested but still could reach the pre-pupal stage when they were parasitized by *D. semiclausum*, whereas host development and metamorphosis were not significantly interfered by *O. sokolowskii*. Similarity analyses of haemolymph proteins of parasitized host larvae proved that the parasitoid's products determine the regulatory ability of parasitoids on host development. Inter-specific competition experiments testified that *C. plutellae* has the strongest ability of parasitism, which partly resulted from the function of teratocytes that also prevent synparasitism from occurrence. The results also showed that different parasitoids have different physiological effects on the host fat bodies. The physiological mechanisms for inter-specific competition of endoparasitoids were discussed in the end.

Ecological and evolutionary patterns in insect metabolic rate and gas exchange

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Metabolic rate is one of the most significant characteristics of organisms. It provides an estimate of the costs of living, and can therefore provide insight into the evolution of life histories across a range of environments. In consequence, much attention has been given to the factors explaining metabolic rate variation, and the way in which gases are exchanged to meet metabolic demands whilst not compromising other aspects of an organism's physiology. Despite this attention, surprisingly little is known about many aspects of variation in insect metabolic rates and gas exchange patterns. Here, we explore ecological and evolutionary patterns in insect metabolic rate and gas exchange. In particular we consider phylogenetic variation in gas exchange patterns and the way in which this variation can be analyzed, environmental (particularly temperature and water availability) correlates of metabolic rate and gas exchange pattern, and body size related variation in metabolic rate.

Identification of isopeptidase activity in the midgut of insects: purification and properties of a *Hofmannophila pseudospretella* (Lepidoptera: Oecophoridae) larval enzyme and possible nutritional ecology

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A g–glutamyl transpeptidase (isopeptidase) has been purified 580-fold to homogeneity from the midgut of keratinophagous larvae of *Hofmannophila pseudospretella*. The enzyme is a single peptide of molecular mass 80 kDa. The enzyme was identified by its hydrolytic activity against the synthetic substrate, g–glutamyl-AMC, its molecular mass and inhibition profile compared to other g–glutamyl transpeptidases and by its fragmentation pattern during mass spectrometry. The enzyme is low or absent from most other insect digestive systems apart from other keratinophagous lepidopteran larvae and predatory carabids. While isopeptide bonds are present in high levels of the proteins in the diet of keratinophages their presence in the diet of predatory beetles has not been established.

Regulation of protein expression of *Heliothis virescens* parasitized by *Toxoneuron nigriceps* at the transcriptional level.

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The endoparasitoid *Toxoneuron nigriceps* has been shown to alter the availability of circulating proteins in the hemolymph of parasitized hosts. Some of the proteins affected are the host storage proteins, and they represent the majority of the proteins available and are key in sustaining processes associated with host metamorphosis. They are also thought to be an important nutritional resource to the developing parasitoid, serving as a source of phenolic amino acids that will be required at the end of the parasitoid larval development. We will report the changes in the level of expression of host proteins involved in developmental processes regulated by this parasitoid, and discuss its implications in the manipulation of the host as a nutritional environment.

DISEASE RESISTANCE IN CROWDS, DENSITY-DEPENDENT PROPHYLAXIS IN THE EGYPTIAN ARMYWORM

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Several insect species display altered phenology and behaviour during outbreaks, a phenomenon known as density dependent phase-polyphenism. The crowded phase is characterised by darkening or melanization of the cuticle, making these individuals more conspicuous than solitary phase individuals. The adaptive value of melanism in the crowded phase is unclear but it may be linked to disease resistance. The density-dependent prophylaxis hypothesis, states that as the risk of parasitism is expected to increase with population density, so species that encounter large fluctuations in density may alter their investment in costly immune defences to match the probability of exposure. Despite growing evidence that insects in high-density populations show increased resistance to certain pathogens, few studies have examined any underlying alteration in immune function. The aim of this study was to quantify variation in immunity associated with solitary and crowded phases in a phase-polyphenic Lepidopteran species (*Spodoptera littoralis*). Relative to pale individuals, melanic larvae exhibited higher haemolymph and cuticular phenoloxidase (PO) activity, and exhibited a stronger encapsulation response to an artificial parasite. Conversely, pale larvae had higher antibacterial activity than melanic larvae. These results are examined in relation to pathogen resistance, and the possibility of a trade-off within the immune system is discussed.

Pharma-entomology: From bugs to drugs

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For decades, natural products, originating largely from plants and microbes, have been a wellspring of new drugs generating more than half of the new drugs registered. However, many pharmaceutical companies have abandoned natural products in favour of mass-produced fully synthetic combinatorial libraries. The expected surge in productivity has not materialized resulting in a marked decline in the output of the R&D programs and, as a consequence, putting a new emphasis on the exploration of biodiversity. Insects represent a major untapped source of novel candidates for drug development. Although they represent the largest and arguably the most successful group of organisms in the world, insects are still largely overlooked as a source of new drug molecules. Insect-derived molecules have a huge variety of potential uses for treating human diseases. From anti-microbial peptides that use completely novel mode of action, to anti-proliferative small molecule drugs, via anti-inflammatory, anti-viral and ion channel modulator molecules, the full scope and breadth available is yet to be realized. Entomed has recognized the promise held by the extraordinary insect chemical versatility and is combining expertise in insect biology, ethno-pharmacology with essential drug discovery and development skills to build the new field of 'pharma-entomology'.

Respiratory strategies used by desert beetles

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This comparative study investigated the different spiracles and forms of respiration used by arid-dwelling beetles to reduce respiratory water loss. The beetles were collected from arid areas within southern Africa and Australia, and possessed a sealed subelytral cavity. Thus, in these beetles, the first pair of spiracles (mesothoracic) opens into the external dry air whereas all the other seven pairs (metathoracic and abdominal) open into the more humid air within the subelytral cavity. Using flow-through respirometry the carbon dioxide emission and oxygen uptake was measured from the mesothoracic spiracles separately and simultaneously with that from around the elytral case. The mesothoracic spiracles were not the main site of gas exchange in all the beetle species, although increased use of these spiracles was associated with increased habitat aridity. The discontinuous gas exchange pattern, another method used by beetles to reduce respiratory water loss, was not observed in all the species. However, most of the species used more convection than diffusion for carbon dioxide emission, regardless of whether the spiracles were closed intermittently or not. The site of oxygen ingress was also determined. These results are explained in the context of elytra shape, food type and activity pattern of the different species.

Dielectric waveguide antenna theory helps explain insect detection of enantiomers

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Many decades ago, a debate arose as to how insects detected semiochemicals. One group maintained that detection was accomplished by electromagnetic energy emanating from the semiochemical, while the other group maintained that the molecule fit into a receptor in order for it to be detected. The latter group's theory is termed the "lock and key model". It is the predominant theory today. The former group's theory was termed the "vibrational theory of odor" by Robert Wright in the 1950's. In the 1960's, Philip Callahan offered a mechanism for this detection whereby the insect sensillae act as dielectric antennae. Behavioral and electrophysiological tests involving the detection of enantiomers could not be easily explained by either theory. Some insects could differentiate between enantiomers while others could not. A more thorough knowledge of antenna theory reveals that enantiomer detection can be explained by the dielectric antenna model. Additionally, replicas of insect sensillae can be artificially reproduced by conventional lasers to make efficient infrared detectors.

Towards an integrative analysis of midgut function in *Helicoverpa armigera*

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CSIRO Entomology has a significant research effort on lepidopteran gut physiology, focussed on digestion, dietary adaptation to natural host plant defence chemistry and the molecular and genetic basis of polyphagy. Our principal target insect is the noctuid *Helicovera armigera* and the project is taking an integrative approach that employs genomic, proteomic and molecular cell biology techniques in conjunction with more traditional physiological methods. The presentation will summarise our recent progress. An Expressed Sequence Tag database was constructed for genes expressed in the larval midgut. From approximately 10,000 clones, a non-redundant gene set consisting of approximately 3900 sequences was identified. This non-redundant gene set has formed the basis for construction of cDNA microarrays that are being used to analyse transcriptional responses to larval feeding on a range of synthetic and natural diets, including chickpea species and *Arabidopsis* ecotypes with differing levels of resistance to *H. armigera* feeding. To complement the genomics-based work, we are concurrently analysing the midgut proteome using 2D-PAGE display, mass spectrometry and functional protein iling. The major classes of digestive hydrolases, including proteinases, carbohydrases and lipases, have been identified from the EST database. The information obtained considerably extends our current understanding of these gene families in lepidopteran insects.

Insects as flying machines

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Insects are small flying machines with an apparent disregard of good aerodynamic practice. Instead of producing lift with their wings, some butterflies rely on ag for slow flight. Tiny insects were predicted to 'swim' through the air using ag, but they rely more on lift. Hovering dragonflies and hoverflies flap their wings in a plane inclined to the horizontal. They use dynamic stall on the downstroke to support their weight, and their lift-to-ag ratio is necessarily less than 2. Most hovering insects flap their wings in a horizontal plane, using lift on both the downstroke and upstroke to support their weight. A leading-edge vortex created by dynamic stall enhances lift production. Because of flow separation at the leading edge, the resultant aerodynamic force is approximately normal to the wing surface, and the lift-to-ag ratio is again typically less than 2. The lift-to-ag ratios for all high-force mechanisms employed by insects are appallingly low by conventional standards, and would entail a high power consumption for fixed and rotary wing designs. For small man-made flying machines to exploit these mechanisms, a flapping motion is essential to ensure that weight is supported by the resultant aerodynamic force and not just the lift.

Caterpillar secretions and induced plant responses: multiple glands mediate plant responses

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Noctuid larvae produce secretions from several glands during feeding. The secretions originate from two separate salivary glands (i.e., labial and mandibular) and from the eversible cervical gland. Each of these secretions may play a significant role in mediating induced responses in their host plants. We have employed a variety of techniques including physical ablation of the structures and genetic ablation (RNAi suppression of genes) to investigate the functional roles of these secretions. The noctuid *Helicoverpa zea* secretes massive amounts of the salivary protein glucose oxidase during feeding on its host plants. We have found that in certain hosts, this enzyme suppresses the induced defenses of the plant, but in other instances may act as an elicitor.

Citrus leafhopper IPM in Queensland, Australia

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Empoasca smithi, is a serious pest of the valuable early varieties Imperial mandarin and Navel orange, and in bad years also of mid- and late-season varieties in Queensland's Central Burnett area. Adults, which breed and feed on flush growth through spring and summer, switch in autumn and winter to green mature and colouring fruit where they cause skin blemishes, downgrading of up to 80% of fruit. Two mymarid wasp parasitoids, *Stethynium empoascae* and *Anagrus baeri*, kill up to 100% of eggs, but need insecticide assistance in most years. Endosulfan at 30 ml/100 L is cheap and effective, killing both nymphs and adults, but must be re-applied to kill hatching nymphs. The insect growth regulator buprofezin kills only nymphs, but also is very effective, though expensive - but contributes significantly to red scale and mealybug control. An orchard-wide strategy and good scouting and decision making is required to prevent re-infestation of treated trees by adults dispersing from hardening flush in untreated trees, especially in late autumn from mid-season varieties with non-susceptible fruit to early season varieties with maturing fruit, and from the common exotic weed tree species *Celtis sinesis*. The benefit to growers of this research is estimated at $11,250-$38,500/ha.

Host-parasite interactions between whiteflies and their parasitoids

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Host-parasite interactions that occur between lepidopterans and their parasitoids are well-documented, but little is known about similar interactions between whiteflies and their parasitoids. Currently, there is no evidence to support the existence of wasp-associated polydnaviruses or venom in redirecting whitefly metabolism, including interference with the immune response. The production of teratocytes by whitefly parasitoids (may play a role in protecting the parasitoid) has been observed for *Encarsia berlesei* and *E. citrina*. Manipulation of hormone titers, so common in parasitized lepidopterans, also occur in parasitized whiteflies. In the *Eretmocerus-Bemisia* system, ecdysteroid titers decrease upon parasitization. However, *Encarsia. formosa* developing in both the greenhouse whitefly (*Trialeurodes vaporariorum*) and the sweet potato whitefly (*Bemisia tabaci*, biotype B) will not molt to its 3rd (last) instar until the whitefly has initiated adult development, a time when host ecdysteroid titers are known to peak. In the *Trialeurodes lauri-Encarsia. scapeata* system in which host and parasitoid enter diapause in May-June, it appears that *E. scapeata* is able to prematurely trigger the initiation of its host's development. These and other host-parasite interactions will be discussed.

Calyx fluid proteins of two biotypes of *Cotesia sesamiae* (Hymenoptera: Braconidae) in Kenya: Implications to biocontrol of *Busseola fusca* (Lepidoptera: Noctuidae)

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Busseola fusca Fuller (Lepidoptera: Noctuidae) is a major stem borer causing damage to cereal crops, especially maize, in most of sub-Saharan Africa. *Cotesia sesamiae* (Cameron) (Hymenoptera: Braconidae) is an indigenous gregarious larval endoparasitoid of *B. fusca* in Kenya and other countries of Africa. There is renewed interest in expanding the geographic range of biocontrol agents of cereal crops in sub-Saharan Africa. One of the targeted agents is *Cotesia sesamiae*, whose importance varies considerably between regions for yet unknown reasons. In Kenya, reports suggest that *C. sesamiae* occurs in two biotypes with different abilities to develop in *Busseola fusca*. A *C. sesamiae* population from coastal Kenya, where *B. fusca* does not occur, does not complete development in *B. fusca* and all its eggs are encapsulated. However, *C. sesamiae* successfully parasitizes *B. fusca* in western Kenya where they occur sympatrically. Recent studies showed that calyx fluid of the two strains is involved in suppression of the immune system of *B. fusca*. Hence the proteins involved in the suppression are genetically different. This study analyses and compares proteins of calyx fluid of these two *C. sesamiae* populations using 2d-Gel electrophoresis and discusses the implications of using *C. sesamiae* as a biocontrol agent in Africa.

Mechanism in the bug *Coreus marginatus* (Heteroptera, Coreidae)

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The structure and function of the hemelytra-to-hindwing locking mechanism of the bug *Coreus marginatus* were analysed. The system consists of a cuticular protrusion in the ventral side of the hemelytra, which locks the subcostal border of the hindwing in flight. The speed and distance slid by both surfaces against one another during flight were assessed using a combination of high-speed video recordings and a 2D geometrical model. The friction coefficient between sliding surfaces was assessed using a micromanipulator, coupled with force transducers. This was done under three experimental conditions: freshly dissected, air dried and rehydrated ethanol preserved samples. The results showed a high speed of sliding, approximately 0.18 $m \cdot s^{-1}$, with a relatively low friction coefficient (0.2 μ). The surface morphology of this mechanism, namely outgrowths of one part having rounded edges, and completely flat surface on the counterpart, effectively aids in the reduction of friction at the microscopical level. The structure is effective even y, and after being preserved in ethanol, suggesting that no cuticle secreted lubrication substance is responsible for its effectiveness. The ultrastructure presumably confers mechanical stability to the system under the high load it is subjected to in flight.

Mandible movements: biting and prey capture

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In many insects, the mandibles are the most powerful mouthparts used for biting, chewing and prey capture. In ants, mandibles are involved in diverse tasks such as manipulating larvae or fungus gardens, grooming, carrying heavy loads, chewing, digging, biting, and predatory strikes. Mandibles generate a wide range of forces and velocities ranging from the softest touch to forceful seed cracking or super-fast strikes. Mandible closer muscles are the largest muscles in the body of ants and are composed of sets of muscle fibers that differ in their morphological and contraction properties. Force and speed output also depend on the arrangement of muscle fibers and apodemes (tendons) within the head capsule, which are reflected by the head shape (long heads = fast, broad heads = forceful). Mandible muscles are supplied by arrays of large motor neurons integrated into sensory feedback circuits that allow the required subtle force control. A particularly fast predatory mandible strike has independently evolved repeatedly in different ant taxa. These 'trap-jaws' are powered by a catapult mechanism and comprise some of the fastest reflexes known.

Respiratory response of insects to hypoxia and anoxia

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The respiratory responses in insects that are highly tolerant to hypoxia may contribute to their adaptation to these environments but are not well understood. We have investigated the respiratory response of individual insects to hypoxia using a sensitive, flow-through CO_2 and water analyser, coupled with an O_2 probe. Respiration rates and patterns in *Tenebrio molitor* larvae exposed to reduced O_2 for 10 min, then returned to normoxia, were monitored. In 10, 5 and 2% atmospheres the normal ventilation patterns of the larvae were completely disrupted although the output of CO_2 was maintained; this pattern suggests the larval ventilation rate had been greatly increased and the spiracles remained open during hypoxia. When the larvae were returned to normoxia, CO_2 release was significantly depressed in the larvae exposed to 5 and 2% O_2 and recovery was slow. Mild hypoxia (15% O_2) had little discernable effect on ventilation pattern or volume of CO_2 released. The respiration in highly tolerant *Sitophilus oryzae* larvae was strongly suppressed during 1 h of anoxia but had recovered to its normal pattern within 10 min of returning to normoxia. Respiratory water loss was minimal in both insects during short exposures to hypoxia despite the increased ventilation rate in *T. molitor*.

Rearing oxygen effects on body and tracheal morphology in insects

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Normal development of the tracheal and body morphology of insects can be influenced by tissue oxygen levels. This oxygen-responsiveness provides a mechanism by which insects can compensate for variation in rearing oxygen level. Despite the many mechanisms of physiological compensation available, micro-electrode measurements in locusts indicate that tissue oxygen levels correlate with air oxygen content. Single-generation exposure of fruitflies to hypoxia or hyperoxia induces compensatory changes in the diameters of the dorsal longitudinal tracheae. Multi-generation exposure to hypoxia or hyperoxia (laboratory natural selection experiments) produces heritable changes in tracheal diameters, indicating that rearing oxygen level can influence the evolution of tracheal dimensions. Body size is positively correlated with rearing oxygen levels in fruitflies for both single-generation and multi-generational experiments, consistent with the notion that atmospheric oxygen can limit insect size. However, effects of atmospheric oxygen level on body size are inconsistent across insect species (mealworms, locusts), so perhaps responsiveness of body size to oxygen depends on phylogenetic history or ecology.

Gregarious development of a solitary endo-parasitoid in its habitual hosts

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Microplitis rufiventris Kok. wasp attacks and can develop on earlier instars of *Spodoptera littoralis* (Boisd.) larvae with preference to third ones. All experiments were carried out using the newly moulted 6th instar larvae (reluctance hosts) at two different temperatures (20±1 and 27±1°C) to test the effect of variable ranges of superparasitization on the developmental interaction between the parasitoid and its host larvae. The effects seem to be dose (no. of eggs + parasitoid factors)-temperature-dependent. In all experiments, neither controls nor stung larvae underwent a larval/larval moult. The parasitoid eggs in singly parasitized host larvae invariably died. However, as the number of parasitoid eggs/host larvae increased, the rate of parasitoid's egg hatching and number of parasitoid larvae successfully reached to their final instar increased. This indicates that the parasitoid has become submissive to the others and stopped fratricide probably due to host's hormone and/or excessive dilution of factors injected with parasitoid eggs. The wasp larvae developed normally to the point of emergence but most did not successfully emerge from the host. The improvement of hatchability and normal post-embryonic development of parasitoid wasp were significantly ($P<0.01$) greater at low temperature (20°C) than at higher one (27°C).

Mechanisms for regulating oxygen delivery in pupae

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Moth pupae are a useful model to investigate the regulation of oxygen uptake in insects. Due to their large size, the uptake of respiratory gases into the tracheal system can be studied directly by small electrodes inserted through single spiracles eventually combined with standard respiratory techniques. We investigated the regulatory capabilities in diapausing moth pupae showing a DGC both under normoxic, hypoxic and hyperoxic conditions. The main part of gas exchange takes place via the spiracles. In order to meet the aerobic metabolic demands of the tissues, moth pupae adjust spiracular conductance by modulation of opening frequency, area and duration. During "microopening", oxygen enters the tracheal system by two mechanisms: convection along a hydrostatic pressure gradient and diffusion, following an partial pressure gradient. We found that oxygen uptake by convection plays a role shortly after the constriction period whereas in the further stage of the fluttering period diffusion plays the main role. Modulating ambient oxygen partial pressure evoked a quick change in spiracular conductance despite of crucial changes in ambient oxygen partial pressure. Under hyperoxic conditions, pupae decreased spiracular conductance by restricting the number of microopenings. The morphological and physiological parameters used to regulate oxygen uptake are discussed in a model.

Contribution of dietary antioxidants (ascorbate and plant phenolics) and low O_2 to the powerful antioxidant capacity of *Manduca sexta* midgut fluid

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Many folivorous caterpillars ingest large quantities of phenolics that can potentially produce reactive by-products, such as semiquinone radicals, hydroxyl radicals and superoxide when oxidized in the midgut. However, the extent to which these actually induce oxidative stress in insects is still debated. In some taxa, oxidative reactions may be minimized by the presence of endogenous or dietary antioxidants or low O_2 availability in the lumen. The midgut fluid of *Manduca sexta* has a powerful antioxidant capacity, and molecular O_2 is rapidly depleted to near-anaerobic conditions. We will present data on the biochemical mechanisms that underlie this phenomenon. We will also show that at least in *Manduca*, ingested phenolics (quercetin, catechin, chlorogenic acid and tannic acid) appear to retain their antioxidant properties (ability to quench a solution of ABTS radical) when mixed with midgut fluid or hemolymph. Caffeic acid exhibits only modest prooxidant properties when mixed midgut fluid. Overall, however, phenolics contribute relatively little to the total antioxidant capacity of the midgut fluid, as demonstrated by comparisons before and after their removal with polyvinylpyrrolidine. The powerful antioxidant nature of midgut fluid is also relatively insensitive to the availability of ascorbate in the diet.

Changes in the haemolymph proteome of the host *Spodoptera littoralis* (Noctuidae) if parasitized by the parasitic wasp *Chelonus inanitus* (Braconidae)

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Successful parasitism of many endoparasitic wasps depends on the supporting action of symbiontic polydnaviruses. *Chelonus inanitus* induces in its host a precocious onset of metamorphosis and a developmental arrest in the prepupal stage. The polydnavirus prevents encapsulation of the parasitoid and causes the developmental arrest, while the parasitoid is responsible for the precocious onset of metamorphosis. Here we report that substances in the haemolymph of polydnavirus/venom containing hosts must be responsible for the developmental arrest. To get an overview of the changes in the protein composition in the haemolymph of non-parasitized, polydnavirus/venom containing and parasitized larvae we analysed the haemolymph proteome by 2D-gel electrophoresis in various stages. In addition, proteins released by the parasitoid larva were analysed. We found that 8 proteins are due to polydnavirus/venom and are analysing them in more detail. 6 proteins are due to the parasitoid larva. Some of the parasitoid released proteins could already be N-terminally sequenced and might play a role in inducing the precocious onset of metamorphosis.

Endoparasitic Strepsiptera masquerade as self in host-derived epidermal bag

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Stichotrema dallatorreanum Hofeneder (Strepsiptera: Myrmecolacidae) manipulates host epidermal tissue and wraps itself within it; which probably camouflages the endoparasitic stages; which probably camouflages the endoparasite and is recognized as ``self'' by the host. This mechanism is one of immune avoidance among parasitoid insects. The host-derived epidermal ``bag'' might have enabled Strepsiptera to radiate to disparate hosts compared with the relatively few taxa (596 species) described so far. They have been recorded as parasitizing 34 families belonging to seven orders of Insecta. While enclosed in the bag an unusual mechanism of insect ecdysis occurs between the first- and second-instar larval stages whereby the cuticle of the 1st instar breaks along the base of the head capsule instead of along the mid dorsal line, as in other insects. The endoparasitic stages undergo apolysis without ecdysis while enclosed within the "bag".

Polydnavirus genome sequencing: exploring polydnavirus structure and gene function through comparative genomics

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Polydnavirus (PDV) sequencing projects have provided novel insights into PDV genome organizations and unique opportunities for exploring viral gene composition and function. Comparisons between the *Campoletis sonorensis* Ichnovirus (*Cs*IV), *Hyposoter fugitivus* Ichnovirus (*Hf*IV) and *Microplitis demolitor* Bracovirus (*Md*BV) genomes have revealed consistencies in aggregate genome sizes and the presence of repetitive non-coding sequence, but have delineated distinct differences in gene composition, viral segment number and topology occurring both within and between the PDV genera. Comparisons between coding sequences within these PDVs indicate the presence of diverse genes and multi-gene families that are unique to individual viral species, conserved within viruses of a given genus, and conserved between viral DNA of both IV and BV origin. Database searches against viral ORFs indicate that many possess significant similarity to known insect genes suggesting that PDVs are accumulating and diversifying invertebrate host genes for pathogenic viral activity. Ongoing analyses of gene and protein expression patterns, sequences, and functions have begun to elucidate that variants within gene families, such as the *Cs*IV *cys-motif* family and the conserved *Cs*IV, *Hf*IV, and *Md*BV *viral ankyrin/I*kb families, may be evolving to play both redundant and unique physiological roles during parasitization.

Balancing oxygen uptake, CO_2 release and water loss in *Periplaneta americana* L.

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An enhanced Krogh model reveals the limitations of continuous O_2 uptake, continuous CO_2 release and H_2O loss at changing oxygen demands: continuous exchanges would be limited by a hyperoxic tracheolar $^{TRL}P_{O2}$ of ca. 15 kPa due to the limited maximal $^{TRL}P_{CO2}$ of 6 kPa. Cockroaches utilize the dangerous $^{TRL}P_{O2}$ as a reserve during bouts of running, walking or preening by CO_2 retention in body buffers in a closing strategy (CS) which also prevents H_2O loss. At the minimal $^{TRL}P_{O2}$ of ca. 1 kPa the accumulated CO_2 is released irregularly by autoventilation. This maximises convective and diffusive O_2 and CO_2 fluxes over the H_2O fluxes as DP_{O2} and DP_{CO2} rise in contrast to the constant DP_{H2O} in a specifically water saving partial pressure strategy (PPS). Resting insects use a combined CS+PPS in CFB cycles where DP_{O2} is maximized in C (closed) periods, and DP_{CO2} is further maximized over DP_{H2O} between repetitive micro-openings in F (flutter) periods in which there is maximal O_2 influx. Adult insects, except ants, use the CS+PPS combination during active ventilation (V) even in the burst (B): in CFV cycles diffusive and convective CO_2/O_2 fluxes are confined to short expiratory/inspiratory openings of the spiracles to retain H_2O specifically.

Characterisation of the digestive system of grape phylloxera (Hemiptera: Phylloxeridae)

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Grape Phylloxera, *Daktulosphaira vitifoliae* Fitch (Hemiptera: Phylloxeridae) feeds on the root system of European grapevine (*Vitis vinifera* L.) leading to the development of galls, vine decline and economic loss to the commercial vigneron. Despite being recognised as a significant pest since the late 1800s, the digestive physiology of phylloxera remains poorly characterised. Previous studies have suggested that the gut is blind ending, although both the presence and absence of an anus has been reported. The structure and function of the insects' digestive system was reinvestigated through the use of modern microscopic (light and scanning electron) and molecular techniques. Phylloxera has an atypical digestive system, with the hindgut emerging dorsal to the midgut. The reproductive system dominates the insects' internal structure. The hindgut intertwines with the ovary duct, emerging dorsal to the gonopore at the posterior end of the insect. Multiple egg development may block the function of the hindgut. Molecular analysis confirmed the absence of bacterial endosymbionts, suggesting that phylloxera is very efficient at converting a high protein diet from individual cells into energy. This research has improved our understanding of phylloxera's physiology. By understanding phylloxera's nutritional requirements, and interactions with grapevines, insect survival and development with changing vineyard management may be predictable.

Immune investment and sexual ornamentation

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The relationship between immune investment and sexual ornamentation is an important subject because insects may trade-off investment in these two costly aspects of their biology. I will describe how ornament size and immune investment covary in two very different systems, the horned dung beetle *Euoniticellus intermedius* and the stalk-eyed fly *Cyrtodiopsis dalmanni*. I will discuss the importance of the 'resource bottleneck' of pupation in these trade-offs, and I will review how our findings relate to sexual selection theory, and in particular immunocompetence handicap ideas.

Genetic dissection of oxygen response pathways and tracheole formation in Drosophila

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We are using genetic and genomic approaches to elucidate the developmental program that governs branching morphogenesis of the *Drosophila* larval tracheal system, a network of ~10,000 interconnected epithelial tubes that transports oxygen to the tissues. These studies have begun to reveal the cellular and molecular processes that induce sprouting and guide outgrowth of tracheal branches and dictate tube size and stability. An FGF ligand (Branchless) guides the migrations of the tracheal epithelium as it grows out and assembles into primary branches. Cells at the ends of the primary branches are induced to express secondary and terminal genes, and go on to form secondary and terminal branches. Primary and secondary branching is highly stereotyped and controlled by a hard-wired developmental program, whereas terminal branch (tracheole) sprouting and outgrowth is highly variable and regulated by tissue oxygen need, as Wigglesworth showed for Rhodnius prolixus 50 years ago. Oxygen control of terminal branching is mediated by the same FGF ligand that controls the earlier, hard-wired stages of branching, but during terminal branching expression of the ligand is regulated by oxygen. We will describe the oxygen response pathway that senses cellular oxygen levels and induces expression of Branchless FGF in oxygen-starved (hypoxic) cells. Secreted Branchless protein activates the FGF receptor Breathless on nearby tracheal terminal cells, triggering a signal transduction cascade that induces expression of tracheole genes and directs cytoplasmic outgrowth to the hypoxic cell. A lumen forms within each cytoplasmic projection, creating new tracheoles that transport oxygen to the hypoxic cell and relieve the oxygen crisis. We will also describe how the same oxygen response pathway also induces a remarkable cellular phenomenon described by Wigglesworth that allows hypoxic cells to locate, capture, and rearrange existing air-filled tracheoles to enhance their oxygen supply.

Immunological adaptation in invertebrates

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Immunological adaptations are central to our understanding of the coevolution between parasites and their hosts, such as invertebrate disease vectors. However, the immune systems of insects and other invertebrates are assumed to have no specific immunological memory, which characterizes acquired immunity in vertebrates. On the other hand, studies of ecological immunity, which demonstrated genotype-specific interactions between invertebrate hosts and their parasites, indicate that invertebrate immunity might be much more complex than is generally believed. We investigated the degree of specificity in the defence of the copepod *Macrocyclops albidus*, against a natural parasite, the tapeworm *Schistocephalus solidus*. We found that the degree of reinfection depends on the antigenic resemblance between consecutively encountered parasites. These results give evidence for specific immune memory in an invertebrate species. Given specific immunity of their hosts, parasites should benefit from antigenic heterogeneity. We are therefore now studying antigenic heterogeneity in *S. solidus*, using fluorescent lectins, which bind specific sugar residues on the parasite surface. We will relate these characteristics to infectivity and innate and adaptive immune activity in the two hosts, copepods and stickleback fishes, to investigate possible constraints for parasites that coevolve with both invertebrate and vertebrate immune systems.

Maternal spread of *Enterobacter agglomerans* and *Klebsiella pneumoniae* through populations of the Mediterranean fruit fly, *Ceratitis capitata* (Weidemann): another piece of the symbiotic puzzle

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Enterobacter agglomerans and *Klebsiella pneumoniae* have been associated with fruit fly pests within four genera of Tephritidae. The associations include aspects of fruit fly nutritional, chemical, and behavioral ecology. The role these bacteria play in the life history of pest tephritids is not fully understood, however recent findings suggest that these bacteria are important in maintaining a healthy and functional gut. Probiotic diet formulations containing *E. agglomerans* and *K. pneumoniae* have been shown to improve the gut of irradiated male *C. capitata* and *A. ludens* used for SIT. In addition, sterile *C. capitata* have demonstrated improved mating efficiency after feeding on particular probiotic formulations. Reports of horizontal and now vertical transmission of *E. agglomerans* and *K. pneumoniae* strongly link these two bacteria to tephritid fitness.

Tracheal capacity and oxygen delivery during flight of Dr*osophila*

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The respiratory system of insects must maximize the flux of respiratory gases through the spiracles of the tracheal system. During flight the supply of oxygen becomes crucial when locomotor activity is increased during maximum aerodynamic force production. In Dr*osophila* most of the gas exchange is due to the 4 large thoracic spiracles that match their opening area to the metabolic need of the animal during flight. To estimate tracheal gas capacity in this insect and to understand the significance of spiracle opening behaviour for oxygen supply of flight muscles we have flown Dr*osophila* in a flight simulator in which we measured carbon dioxide release and locomotor performance during visually controlled flight. By blocking various spiracle openings for gas exchange we varied oxygen supply and measured the changes in muscle mechanical power output delivered to each wing including muscle efficiency. Data show that a reduction of spiracle gas exchange area impairs maximum locomotor capacity that results in a decrease in stroke amplitude and aerodynamic force production. The release of carbon dioxide tends to be more cyclic under these experimental conditions suggesting that the insect might employ discontinuous breathing patterns even at elevated locomotor activity.

Parasitoid wasp uses a venom cocktail injected into the brain to manipulate the behavior and metabolism of its cockroach prey

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Parasitoid wasps incapacitate their prey to provide a food supply for their offspring. Because the wasp's goal is to provide a living meal for its newborn larva, the behavioral and metabolic changes in the host are brought about by manipulating the host behavior and metabolism in a way beneficial to the wasp and its offspring. To this end, these wasps have evolved venom-cocktails tailored to the wasp hunting and its larva feeding strategies. The two consecutive stings of the parasitoid wasp *Ampulex compressa* are unusual, as they induce a transient paralysis of the front legs followed by grooming behavior and then by a long-term hypokinesia of the cockroach prey. The first sting in the thoracic central nervous system causes a transient front leg paralysis lasting a few minutes. This paralysis is due to the presence of a venom component that induces a postsynaptic block of central cholinergic synaptic transmission. Following the head sting directly into the brain, dopamine identified in the venom appears to induce 30 minutes of intense grooming. During the long-term hypokinesia that follows the grooming, specific behaviors of the prey are inhibited while others are unaffected and the overall metabolic rate of the cockroach is modulated.

Insect midgut proteinases

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Most initial protein digestion takes place at the endoperitrophic space in insect midguts and depends on three classes of proteinases: aspartic, cysteine and serine proteinases. Aspartic proteinases are described in some few insects and are preferentially active on substrates containing a Phe-Phe peptide bond. Cysteine proteinases occur in Hemiptera and some Coleoptera as *Tenebrio molitor* and are active on B-R-pNA, B-R-NA, Z-FR-MCA and e-amino-caproyl-leucyl-(S-benzyl)-cysteinyl-MCA are activated by DTT and cysteine and are inhibited by E-64. Five cysteine proteinase cDNAs were sequenced and cloned from a midgut cDNA library from *T. molitor* larvae: three lysosomal, one digestive and another probably digestive. A Neighbor-Joining (NJ) analysis of all cysteine-proteinase sequences from different insect species described in GenBank distinguish two main branches: one which contains typical digestive cysteine-proteinases presented by Coleoptera and another with Hemipteran cysteine proteinases closest to lysosomal ones. Serine endopeptidases are the main enzymes involved in protein digestion in insects, usually comprising trypsin and chymotrypsin. Purification and characterization of these enzymes from different insect species revealed enzymes with different specificities and physical properties. Sequence alignment, structure superposition, NJ analysis and molecular modeling allowed the identification of residues involved in resistance to plant metabolites like proteinase inhibitors and ketones. Supported by: FAPESP, CNPq and PRONEX.

Irritable bowel syndrome in insects: Plant defenses that disrupt the peritrophic matrix

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We have demonstrated that a unique 33-kD cysteine protease (Mir1-CP) accumulates in the whorls of caterpillar-resistant corn genotypes in response to feeding by fall armyworm (*Spodoptera frugiperda*) larvae and other lepidopterans. The growth of fall armyworm larvae reared on transgenic Black Mexican Sweet (BMS) cells that ectopically expressed Mir-CP was retarded approximately 70%. Physiological indices suggested that this was due to impaired nutrient utilization. Scanning electron microscopy (SEM) of midguts from fall armyworm larvae reared on resistant plants or transgenic BMS indicated that peritrophic matrix (PM) was severely damaged. Because Mir1-CP binds chitin, we speculate that it might be tethered to the PM where it digests PM proteins. Proteomic analysis of PM proteins isolated from larvae reared on resistant and susceptible corn genotypes indicated that approximately three proteins were absent from the PM of larvae reared on resistant plants. This suggests that the protease may affect the integrity of the PM by attacking specific PM proteins. To test this Mir1-CP has been expressed in a heterologous expression system using a recombinant baculovirus vector and *Trichoplusia ni* larvae as hosts. Mir1-CP has been purified to homogeneity from the hemolymph and its affects on PM structure will be tested *in vivo* and *in vitro*.

Cellular differentiation and functional development of the organs responsible for sustained active water vapour absorption (WVA) in the firebrat, *Thermobia domestica* (Packard).

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WVA commences sporadically in the second instar but then develops rapidly, reaching its mass-specific peak in late-third and fourth instars before settling to a slightly lower plateau. The dorsal and two ventro-lateral posterior rectal (anal) sacs responsible for WVA develop as outpushings of the proctodaeal invagination. Functionally mature sac epithelial cells are characterised by a very regular, dense honeycomb-like array of elongated mitochondria closely associated with deep pleated infolds of the portasome-studded apical plasma membrane. In the first instar, the prefunctional single-cell thick epithelium convolutes, forming saccules. The epicuticle lifts away from the epithelial surface and traverses only the neck of each sac, with a faintly granular subcuticular matrix filling the expanding intervening space. Mitochondria are manufactured and packed very densely in mitochondrial factory cells. In the second instar, the apical association of mitochondria and plasma membrane begins to appear. By late in the third instar, the fully mature sac epithelium has developed, coinciding with peak WVA. During moults, WVA temporarily ceases as the sac cells dedifferentiate, divide and redifferentiate. A new cuticle is secreted. Mitochondria multiply. When WVA restarts immediately after ecdysis the functionally mature state of the sac cells has been fully re-established.

Cold tolerance and feeding behaviour in the Tasmanian scorpionfly *Apteropanorpa* Carpenter (Mecoptera: Apteropanorpidae)

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The Apteropanorpidae is a small family of scorpion flies endemic to Tasmania, comprising two described species. The biology of the family is almost completely unknown. Adults of both species are found on shrubs in the alpine zone and can be active in winter, although the peak in adult abundance is autumn. Experiments cooling individuals from 3ºC to −10ºC at 1ºC/3 mins and measuring body temperature using infrared video thermography show that live *Apteropanorpa* freeze at an average temperature of −4.9ºC (n=20). However, stress response behaviour is triggered at an average temperature of −1.1ºC (n=20), and no individuals survived freezing to −10ºC. Combined behavioural and physiological data indicate that *Apteropanorpa* does not possess a physiological mechanism to survive freezing, and instead employs freeze avoidance behaviour as the insect seeks microhabitats with favourable temperatures. *Apteropanorpa* is apparently well-adapted to its natural environment; the maritime climate of the Tasmanian alpine zone is relatively warm and stable compared to that of mainland Australia. Adults are most likely saprophagous in nature, predominantly feeding on dead and decaying invertebrates. Thirty-three adults were included in a feeding choice experiment in which twenty-two adults showed a statistically significant preference for feeding on dead arthropods. A saprophagous feeding strategy in the Apteropanorpidae is supported by field observations, histological tests and gut contents analyses by light and electron microscopy.

Geographic variation in host selection behaviour in the *Drosophila* parasitoid *Leptopilina clavipes*

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Coevolutionary processes between hosts and parasitoids are not simply the result from host-parasitoid interactions between two species, but rather are determined by the diffuse interactions between a number of biotic or abiotic factors. An important factor influencing this coevolutionary process is the predictability of the environment. Modelling studies show that in a stable predictable environment, the optimal strategy for the parasitoid is to specialise on the most frequently encountered species. In a fluctuating environment however, high temporal variation in the frequency of different host species do not favour the evolution of specific virulence in the parasitoid. To test this hypothesis, we compared survival probabilities of two strains of *Leptopilina clavipes* a parasitoid of *Drosophila* larvae breeding in fungi. One strain originates from northwestern Europe, where fungi are almost continuously available during the season, the other strain originates from southern Europe, where fungi are absent during an important part of the season. The availability of fungi presents European *L. clavipes* populations with environments that differ in their predictability. We tested survival and host selection behaviour on three different host species from different breeding substrates. Our results confirm the model predictions and show specialisation in northwestern, and generalisation in southern populations.

Host regulation by ***Toxoneuron nigriceps*** **bracovirus genes**

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Toxoneuron nigriceps is an endophagous parasitoid of the tobacco budworm, *Heliothis virescens*. Parasitized host larvae show the suppression of the immune response and are unable to attain the pupal stage, becoming developmentally arrested as mature larvae. This complex syndrome is largely induced by a symbiotic bracovirus (TnBV), which is injected by the adult wasps at the oviposition. The sequence of TnBV genome is in progress and several genes expressed in parasitized hosts have been cloned and fully characterized. Among them, some code for members of interesting protein families, such as IκB-like and tyrosine phosphatases, while others do not show significant levels of sequence identity with known proteins. The current study of these TnBV genes provides new insights on the possible regulation mechanisms of host immune disruption and developmental arrest modulated by *T. nigriceps* parasitism, and shows the occurrence of interesting cellular and molecular pathways of host disguise which seems to occur also in other host-parasitoid systems.

Is reproduction of the silkworm parasitoid *exorista sorbillans* **(diptera: tachinidae) totally dependent on the endosymbiont** *wolbachia?*

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Several species of arthropods harbour endosymbiotic bacteria of the genus *Wolbachia*. These endosymbionts are transmitted vertically from one generation to the next and are facultative / obligatory in several Dipterans that have been studied to date. The tachinid uzifly, *Exorista sorbillans* known to cause extensive damage to silk industry by adopting obligate parasitic mode of life on its host silkworm, *Bombyx mori* L (Lepidoptera: Bombycidae). Further this pest is known that it harbour *Wolbachia* endosymbiont. Administration of 0.05 mg/ml oxytetracycline to the adult uzifly, removed *Wolbachia* endosymbiont and results in causing different invasive effects such as i) reduction in fecundity of aposymbiotic females ii). Cytoplasmic Incompatibility in the crosses between symbiotic male and aposymbiotic female. iii) Partial Genomic incompatibility in crosses between both sexes untreated and in crosses between aposymbiotic males and symbiotic females, as well as in the crosses between both males and females administered with antibiotics. It is hypothesised that, there is a driving force which causes partial genomic incompatibility in crosses between both sexes untreated (uncured) and in crosses between cured male and uncured female, is pronounced might be due to the lack of *Wolbachia*. However, the tetracycline has no much effect on longevity and survivability of uzifly. These results suggest that, *Wolbachia* present in the uzifly display obligate mutualism as it controls the overall development of reproductive physiology of its hosts.

Parasitoid-induced changes in gene expression patterns in *Ephestia kuehniella*

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Parasitism by endoparasitic wasps may significantly alter the host's physiology and ontogeny, including development, growth rates, and immune response. To understand the basic molecular mechanisms underlying these parasitoid-host-interactions and to obtain a first general idea on the genes involved in these processes we have analysed patterns of gene expression by cDNA-AFLP (cDNA-Amplified Fragment Length Polymorphism) in *Ephestia kuehniella* (Lepidoptera: Pyralidae) caterpillars after parasitization by the wasp *Venturia canescens* (Hymenoptera: Ichneumonidae). Comparison of expression patterns of approximately 1860 cDNA fragments in parasitized and unparasitized *E. kuehniella* caterpillars showed that about 1% of the transcripts were suppressed after parasitization, which was further confirmed by virtual Northern blot or RT-PCR analysis. Sequence analysis of the cDNA-AFLP fragments revealed that the putative translated proteins are associated with energy metabolism, protein synthesis, signal transduction, or are part of different membrane proteins. In addition a number of fragments were derived from novel genes not matching any database entries. Our transcriptomic approach therefore demonstrates that parasitization by an endoparasitic wasp changes key developmental processes as well as expression of new specific genes in the lepidopteran host caterpillar.

Genome sequencing of the *Cotesia flavipes* polydnavirus

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Diatraea saccharalis is a key pest of sugarcane, but a minor pest of sweet corn, rice, and sorghum. Natural enemies of *D. saccharalis* have been reported to be important in reducing its populations, thus minimizing damage to crops. *Cotesia flavipes* is a gregarious parasitoid of *D. saccharalis*. The parasitoid injects polydnavirus (PDV) genes into *D. saccharalis*, which are transcribed into proteins that alter the physiology and suppress the immune system of the host. This paper reports the establishment of a *D. saccharalis-C. flavipes* colony, which is being used to construct a genomic library for the *C. flavipes* PDV. The genomic sequencing of *C. flavipes* PDV is under way using the shot gun strategy. The final goal is to identify potential opening reading frames of immune-suppressive genes and others with significant physiological functions.

Ecological adaptation of circadian activity rhythms: Phase shift and loss of rhythmicity with the altitude change in plusinae (Noctuidae) moths

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Most of the Plusinae moths are nocturnal and the rhythms are driven by endogenous circadian rhythmicity. But it is also well known that the silver Y, Autographa gamma shows diurnal migration flight and Alpine species visits flowers in the daytime. We investigated the details of their rhythms using actographs. The Alpine Syngrapha nyiwonis was found to be essentially nocturnal but some, especially mated females, take flights in the daytime. Interestingly, their nocturnal activity is strictly limited in a few hours right after sunset, and they show no circadian component in the free-running state under DD or LL condition. Both of these natures seem to be adaptive to cool and unstable alpine weather. Rhythms of non- and sub-Alpine species are also discussed.

On the immunological memory of insects

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There are several hypotheses as to why insects should not have evolved an immunological memory comparable to that found in the higher vertebrates. Nevertheless, a number of studies suggest that a memory exists - with properties not unlike those of vertebrates. In the light of evolutionary ecology, the issue of a memory cannot be addressed without taking into account the problem of specificity in the immune response. Here, the existing evidence concerning these two questions is discussed and structured within a more general defence framework. Finally, a look at the temporal dynamics of the immune response hints at "solutions" that insects may have found for the problem of immunological memory.

Lectin-induced immune-suppression in insects

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Many insect parasitoids that deposit their eggs inside immature stages of other insect species inactivate the cellular host defence to protect the growing embryo from encapsulation. Suppression of encapsulation by polydnaviruses-encoded immune-suppressors is correlated with specific alterations in hemocytes that include cytoskeletal rearrangements, and the loss of ability to spread on foreign surfaces. We have previously shown that the *Cotesia rubecula* polydnavirus gene product CrV1 is sufficient to cause immune suppression when injected into the host hemocoel, where CrV1 is taken up by hemocytes. Uptake is dependent on dimer formation, which is also a prerequisite for lipophorin binding, an indication that CrV1-lipophorin complexes are involved in hemocyte uptake. Since CrV1 resembles oligomeric lectins regarding interaction with lipophorin and uptake in resting cells, we examined the cytoskeleton during lectin-mediated uptake reactions and observed F-actin depolymerization resembling cytochalasin D inactivation. These observations suggest that some oligomeric adhesion molecules, which may include immune-suppressors, internalise receptors from the cell surface and in the process depolymerize actin-cytoskeleton. Although other more complicated mechanisms are possible, cellular immune inactivation in insect parasitoids may involve massive uptake reactions driven by lipoprotein complexes, which destabilize actin-cytoskeleton. Since recycling of membrane-vesicle to the periphery requires actin-cables, consecutive uptake reactions deplete receptors from the cell surface.

Variation in tracheal and spiracle morphology among insects

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Tracheal systems enable insects to support high aerobic metabolic rates by a direct transport of oxygen to the mitochondria. But organs differ in oxygen demand and the high-capacity respiratory system may cause water loss and excess tissue oxygen levels. Physiology and morphology of tracheae and spiracles must therefore be finely regulated. Thus it can be hypothesized that gross morphology and ultrastructure of tracheal systems and spiracles correlate with the metabolic demands of a species, a developmental stage or a single organ. This hypothesis is tested looking at the tracheal systems and spiracles of insects of different environments and with different metabolic rates. Two examples of quantitative studies of tracheal systems are presented: (i) The tracheal system of the stick insect *Carausius morosus* is an example for a simple system in which air sacs are lacking and organs are hardly penetrated. Nevertheless, the distal fine tracheae are most important in oxygen delivery. (ii) The tracheal gills of the aquatic larvae of the dragonfly *Aeshna cyanea* are an example for the adaptation of tracheal systems to water as respiratory medium. The tracheolar supply of the respiratory epithelia is designed for effective oxygen uptake, supported by ventilation of the respiratory chamber.

Friendly-fire on the battlefield: how important is autoimmunity in insects?

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Insects express a range of immune effector systems in their haemocoel during systemic immune insults. Some of these effector systems function by generating generic cytotoxins in the haemolymph in the vicinity of the pathogen. In theory expression of these effector systems should have autoimmune consequences, and such consequences are a major assumption in many studies of immune costs in ecological immunity. We present the results from experimental tests of this notion and document the autoimmune consequences of expressing the phenoloxidase cascade in response to a controlled immune insult.

Suppression of host cellular defense responses by *Microplitis demolitor* bracovirus

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Host insects often kill parasitoids by a cellular immune response called encapsulation. During an encapsulation response, insect blood cells (hemocytes) attach to the target and form a multilayered sheath. Since hemocytes normally circulate freely in the insect's body cavity (hemocoel), encapsulation requires that hemocytes recognize the target as foreign and change from nonadhesive cells to adhesive cells that attach to the target and one another. Polydnavirus-carrying parasitoids often evade encapsulation by disrupting the host's encapsulation response. Our studies have identified several key factors that regulate capsule formation by the moth *Pseudoplusia includens* and have also determined that *Microplitis demolitor* bracovirus (MdBV) blocks encapsulation by inhibiting infected hemocytes from adhering to foreign surfaces. The MdBV genome encodes several gene families expressed in host hemocytes and other tissues. In this presentation, we discuss the role of selected MdBV genes in suppression of encapsulation and other host immune responses. We also discuss the use of RNA interference (RNAi) and gain of function methods in studying PDV gene function.

Controlling flapping flight: lessons from locusts, dragonflies and *Aedes* mosquitoes.

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To a flying insect, unsteadiness is everything. With wings flapping tens or hundreds of times every second, the airflow changes continually, but despite – or perhaps because of – this complexity, insects are able to control manoeuvres with greater finesse than even the highest performance fighter aircraft. By adopting and adapting techniques used by engineers to analyse aircraft stability and control, it is possible to predict an insect's response characteristics to accidental disturbances and deliberate control inputs. By analysing the insect's natural modes of motion, we can then make quantitative predictions about the operating bandwidth of the nervous system and even the preferred directions and temporal resolution of its sensors. Besides yielding integrative physiological insight, analysing the insect as a miniature flying machine is instructive for the engineer seeking to mimic aspects of insect flight. In this talk I will illustrate some of the lessons we can learn from behaviours as diverse as the cruising flight of locusts, the take-off manoeuvres of dragonflies, and the mating flights of *Aedes* mosquitoes. Finally, I will show how an understanding of flight control can even contribute to pest control, by helping us select the most successful male mosquitoes for use with the sterile insect technique.

Molecular cloning and expression patterns of the molt-regulating transcription factor, Hhr3 from *Helicoverpa armigera*

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A 1668 bp cDNA encoding a molt-regulating transcription factor, hormone receptor 3 (HHR3) was cloned from *H. armigera,* which deduced a protein with 556 amino acids. The isoelectric point of this protein was pI 6.52 and the molecular mass was 62 kDa. A DNA-binding region signature of nuclear hormone receptor was found from No. 107 to 133 amino acids. A possible transmembrane helice from outside to inside was found from No. 72 to 90 amino acids. Five bands of HHR3 were detected from the larvae by northern blotting. Among of them, band 1 and 2 were the dominant populations. The expression patterns of HHR3 *in vivo* were variable upon developmental stages and tissues. Results suggested that bands of 1 4 of HHR3 was only shortly expressed during molting which suggested that they were involved in the regulation of molting cascade, whereas, band 0 was observed in both molting and feeding larvae. Band 1 and 2 of HHR3 could be inducted from epidermis by RH-2485. The cDNA of HHR3 was expressed in *E. col.* A reconstructed HHR3-NPV was prepared and the efficacy to pest insect was studied. The work was supported by grants: 3008002,30330070 and 2001AA214031-1-2.

Immune-related genomics and proteomics of the lepidopteran *Galleria mellonella*

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Screening for and characterization of immune-related proteins from the greater wax moth *Galleria mellonella* resulted in the identification of antimicrobial peptides which have been conserved in bilaterian evolution and in the discovery of novel proteinase inhibitors contributing to innate immunity of this insect. Using the subtractive hybridization in combination with suppressive PCR, we analysed genes that are differentially expressed in haemocytes in response to injected microbial elicitors such as LPS. Among the differentially expressed transcripts we identified homologues of pattern recognition molecules, antibacterial peptides and a novel member of the defensin-like peptides exhibiting antifungal activity which was named Gallerimycin. Application of proteomic methods in targeted screening for immune-related proteins in the haemolmph of preinjected *G. mellonella* larvae resulted in characterization of three inducible serine proteinase inhibitors (ISPI-1-3) which are active against the serine proteinases produced by the entomopathogenic fungus *Metarhizium anisopliae*. In addition, we discovered a specific inhibitor of microbial metalloproteinases (IMPI) which represents the first non-TIMP-like inhibitor of metalloproteinases reported from animals. Our research focussed on non-self recognition and immune-related signal transduction in *G. mellonella* provides evidence that pattern recognition of microbial characters in this insect comprises recognition and regulation of thermolysin-like metalloproteinases associated with invading pathogens.

Intratracheal pressure gradients and oxygen supply in resting and flying hoverflies

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Hoverflies were chosen as extremely steady and rapid fliers in expectation of a very effective respiratory mechanism. The Syrphidae *Eristalis*, *Volucella* and *Sericomyia* were analyzed using non-invasive sensor techniques. At rest, the tracheal system is ventilated by hemolymph shift due to periodic heartbeat reversal causing fluctuations in intratracheal pressure and oxygen content. During shivering and flight, rate of pressure pulses in the thoracic tracheae is identical with wingbeat frequency. While during shivering oxygen content decreases dramatically, during flight oxygen content increases and reaches almost concentrations of ambient air. This reminds of the unidirectional air supply by the flight apparatus in hawkmoths (Wasserthal JEB 204, 2001): During full flight, a pressure gradient arises with a mean negative pressure at the anterior spiracles and a mean positive pressure at the second thoracic spiracles. Downstroke of the wings coincides with inspiration through the first spiracles, upstroke with expiration through the 2nd spiracles. As a unique feature for forced expiratory air flow the 2nd spiracles of *Eristalis* possess a motile filter apparatus combined with the inner valve.

Applications of atomic force microscopy for entomological studies

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The atomic force microscope (AFM) is a relatively new form of microscopy based on the detection of small forces acting between a sharp probe and a surface. The microscope has a spatial resolution of less than a nanometre and can provide a 3 dimensional interaction profile of the probe and sample at the pico-newton level in real time and space. In its most common form of operation the AFM will provide a topographical view of a surface from the micron down to the nano-metre scale. On flat well-ordered surfaces atomic and molecular scale resolution is possible. Unlike the traditional forms of high resolution microscopy (e.g. Scanning electron microscopes) the samples can be good conductors or insulators and thus sample coating is not a requirement. Moreover the samples can be examined in ambient conditions. As the instrument operates effectively in vacuum, air and liquid environments the samples in many cases can be viewed in-situ in a living and functioning state.

Sex-biased immunity and parasitism in insects

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Recent studies of vertebrates, especially mammals and birds, have indicated that males generally have weaker immune systems and are generally more heavily parasitised than females. One mechanistic explanation for sex-biased immunity and parasitism involves the immunodepressive effects of androgen, testosterone. Since insects lack this hormone, they provide a suitable taxon for examining the ubiquity of sex differences in parasitism and immunity, without the confounding influence of testosterone. Here, we report the results of a comparative analysis aimed at determining the direction and magnitude of sex-biased parasitism and immunity in insects and examine some of the proximate mechanisms underpinning the observed relationships.

Tolerance of *Drosphilla melanogaster* to very low levels of oxygen.

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Paper not available at time of production.

Effect on native *Pieris* Butterflies in New England, USA, by *Cotesia glomerata* and *C. rubecula* (Braconidae), Introduced Biological Control Agents of *Pieris rapae*.

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In New England (northeastern USA), *Pieris rapae* is an invasive butterfly (ca 1860), against which two braconid parasitoids were introduced: *Cotesia glomerata* (1884) and *Cotesia rubecula* (1988). Neither introduction was preceded by formal host range estimation. Two butterfly species are at potential risk in the region: *Pieris virginiensis* and *Pieris napi*. Recent studies have shown that both are physiologically acceptable hosts for both parasitoids. Field studies showed that the univoltine woodland nature of *P. virginiensis* protects it from attack by either parasitoid. In contrast, a range reduction of *P. napi* appeared to occur shortly after the *P. rapae* invasion. Currently, critical host plants are abundant in both areas retaining *P. napi* (northern New England) and areas now lacking the species (southern New England), and host plants do not explain this range change. Parasitism by *C. glomerata* is high (66-100%) in the summer generation of *P. napi* in both areas retaining and areas lacking the butterfly. The critical difference appears to be a higher rate of commitment to first generation pupal diapause by caterpillars in wooded habitats in northern New England and absence of these parasitoids in wooded areas has protected northern *P. napi* populations, which has avoided parasitism by effectively becoming univoltine in a parasi

The genetic components of community structure and ecosystem processes: conservation implications

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The heritable genetic variation within individual species, especially dominant and keystone species, has community and ecosystem consequences. These consequences represent extended phenotypes, i.e., the effects of genes at levels higher than the population. Using diverse examples from microbes to vertebrates, the extended phenotype can be traced from the individuals possessing the trait, to the community, and to ecosystem processes such as leaf litter decomposition and N mineralization. Common garden experiments show a heritable component to community structure and ecosystem processes. Our studies also argue that there are genetic based assembly rules that can be used to predict community structure. These findings have important basic and applied implications. For example, the loss of genetic diversity in a dominant tree can result in the loss of community members that are dependent upon specific tree genotypes for their survival. Also, the extended phenotypes of genetically modified organisms can have unexpected community and ecosystem consequences.

A modern metaphor for island biogeography of SW Pacific Simuliidae - the Indiana Jones Zone.

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The simuliid subgenus *Hebridosimulium* from Vanuatu and Fiji, originally thought to be a taxonomically isolated taxon, can now be related to *Wallacellum*, a subgenus sporadically distributed west of New Guinea, but mainly in the Philippines. Simuliid taxa on New Guinea and the Solomon Islands are apparently not closely related to *Hebridosimulium*. The paleogeology of New Guinea, the Solomon Islands, Vanuatu and Fiji shows the region to be of a geological complexity second-to-none in the world. In Fiji, Viti Levu has been dated to some 40 Mya, but other islands only of 7-2 Mya. While some islands of Vanuatu may have been emplaced for some 30+ My, all appear to have been submerged at one time and some more than once, albeit all are presently rising. Colonization of islands that may, or may not, become submerged is similar to the problem presented to Indiana Jones crossing the floor of the Temple of the Holy Grail. Step on (colonize) the wrong tile (island) and Indy falls through the floor (submerges) and cannot move on to reach the Holy Grail (Fiji).

The fine-scale physical attributes of coarse woody debris associated with utilization by ants (Hymenoptera: Formicidae).

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"Coarse woody debris (CWD) is increasingly recognized in Canada for its contribution toward biodiversity. [U1]It is a particularly vital resource in sub-boreal forests as nesting habitat for ants (Formicidae). Wood, which has low specific heat, provides a thermally favourable environment in this cool climate. Ants contribute to physical breakdown of wood, and colonies are a significant food source for vertebrates like grizzly bears and woodpeckers. Where CWD retention has been a management objective, volume is usually used as the sole indicator. However, CWD differs greatly in quality between non-harvested and harvested sites. Pieces remaining after harvest are shorter, thinner, lacking in bark, and missing decay classes typical of non-harvested sites. This has led to the concern that post-harvest CWD may not be biologically functional. In this study, conducted in cool moist sub-boreal forests, we are examining the fine-scale physical attributes of CWD utilized by ants, in non-harvested and harvested sites (8-10 years post-harvest). Results to date indicate significant effects of stand structure and climate on ant ulitization of CWD, with only a few species tolerating cool interior stand conditions. The possible significance of regional climate, insolation, temperature, and moisture will be discussed.
"

Indicators, Flagships and Predictor Sets: Insects as Surrogates

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Invertebrates have been used as indicators of environmental qualities for over fifty years. There remains a lack of clarity of their use in this connection which may lead to inappropriate choices of what to use as an indicator or even of the state of the quantity being indicated. Single species may indicate the quality of their chemical environment when they interact physiologically with that chemistry. In contrast complex ecological changes are unlikely to be reflected by the state of single species. A whole biota will respond to such changes and an indicator for such a complex entity will be, inevitably, a set of species. How we choose such a set is also an issue that requires careful thought. A predictor set acts as a surrogate for the complete biota. Such a set is better chosen for its ecological variability and representativeness than simply for taxonomic convenience. A predictor set of insects is more likely to be effective if it spans different feeding guilds. Examples will be presented and discussed. Flagship species, in contrast, are chosen and used for socio-political purposes and may indicate nothing more than a particular place, time or mind-set.

The concept of effective community size for furthering the development of community genetics

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Community genetics represents the union of community ecology with population and quantitative genetics. This unique perspective emphasizes investigating the influence of genetic variation on interactions between species. Determining when this perspective is important is the subject of ongoing research. In this light, we introduce the concept of "effective community size," the fitness relevant community that an individual experiences. This value represents the weighted number of species' genomes that influence fitness of an individual, and is determined the frequency and strength of interaction of an individual with all other species in its community. We apply this concept to explore two major conditions that are crucial for broad application of a community genetic approach: (1) fitness in one species must covary with genes in a second species and (2) the community context is appropriate such that the effective community size of each interacting species is low. Using simulated communities with controlled, genetically determined species interactions, a community genetic framework breaks down as we increase the effective community size of the participants. We use a well-studied, cottonwood-arthropod system to show that a constant, strong selective influence of dominant or keystone species greatly reduces effective community size of many other species in a community and enhances the need for a community genetics perspective. Recent analyses of community webs from other systems indicate that a large proportion species interact with relatively few other species in a community. Applying the concept of effective community size to these analyses suggests that community genetics is an important perspective in many communities.

Genetic variation in leaf chemistry influences ecosystem processes during decomposition.

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Many tree species display large variation in genetically-mediated leaf chemistries important to insect herbivore defense. These secondary compounds, especially polyphenolics, are also important to ecosystem level processes during decomposition. As natural plant population sizes decrease, genetic diversity within a single species also declines, yet the importance of this loss in genetic variation to ecosystem processes is relatively unknown. We use two model species, *Quercus laevis* and *Populus tremuloides* to demonstrate the ecosystem level effects of intraspecific genetic variation in leaf chemistries. In a three-year field experiment, we varied the intraspecific leaf litter composition of *Q. laevis* in an oak sandhills community and monitored ecosystem responses. Intraspecific variation affected carbon and nitrogen cycles during decomposition through genetically-mediated litter chemistries. Mass loss, changes in litter carbon and nitrogen content, as well as soil ammonium availability were all affected by intraspecific litter composition. In a parallel laboratory microcosm experiment, we tested the effects of intraspecific litter diversity and nitrogen deposition on ecosystem processes. Intraspecific diversity increased soil respiration overall, with the greatest increases in respiration occurring under high nitrogen deposition. In a second field experiment, we also varied the genetic composition of *P. tremuloides* litter grown under different nutrient availability regimes. Decomposition rates varied two-fold within identical nutrient treatments, depending upon the genetic identity of the leaf litter and corresponding litter chemistry, and strong gene by environment effects were evident. While leaf secondary chemistries have traditionally been known to be important to insect herbivore ecology, it is becoming increasingly apparent that they also play important roles in ecosystem level carbon and nitrogen cycling. Our results demonstrate that genetic variation within a single species can have independent, and interactive effects with nutrient availability, to influence terrestrial nutrient cycles.

Conservation of species at the edge: Hybrid zone dynamics and cryptic mountain species impacteded by changing thermal landscapes.

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Hybrid animals may harbor significant genetic novelties, and provide valuable genotypes needed for rapidly changing environments. Recently (and probably directly related to extensive regional global warming in the region since 1998) considerable northward and rapid interspecific introgression (150-350 km in 6 years) into historical *P. canadensis* populations has been observed for several autosomal species-diagnostic traits of *P. glaucus* such as detoxification of tulip tree leaves, morphological wing traits, and *Hk*-100 allozymes. In addition, some X-linked traits such as *Pgd*-100 alleles and tulip tree oviposition preference behavior have moved extensively northward. However, there has been little introgression of certain other sex-linked *P. glaucus* traits (such as facultative diapause, dark morph females, and LDH-100 allozymes). We have determined that recombination and strong selection on *Ldh*-100 alleles (or some undetermined trait on the X-chromosome with very close linkage) seem to limit gene flow North of this interspecific hybrid zone wherever fewer than 2700 degree-days accrue. We have created recombined/introgressed hybrids that largely match the morphological and delayed post-diapause emergences of the new mountain swallowtail species, *P. appalachiensis* (Pavulaan & Wright 2002). The new species expresses an extensive introgressed mix of both *glaucus* and *canadensis* diagnostic traits and may represent the high altitude version of our high latitude northern "false second generation" populations. This neospecies may represent a rare example of an animal "hybrid species". Detailed GIS mapping of thermal accumulations allows prediction of the geographic location of the purportedly new mountain species in West Virginia as well as for Georgia in the southern Appalachian Mts. The exceptionally warm 2002 and 2003 seasons may extirpate the neospecies as genetic swamping from *P. glaucus* may occur from lower elevations.

Eucalypt hybridisation and heteroblasty affect biodiversity of dependent communities

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The relative effects of host hybridisation and ontogeny on the composition and diversity of dependent communities was examined using manipulated hybrids between two heteroblastic forest tree taxa, *Eucalyptus nitens* and *E. globulus.* The abundance of 44 dependent arthropod and fungal taxa was assessed on 3 year old trees of *Eucalyptus globulus*, *E. nitens* and their hybrids (F_1, F_2, and backcrosses) growing in a replicated field trial on the island of Tasmania. Half of the trees of each cross type had undergone the transition to adult foliage (heteroblastic) while the remainder still retained juvenile foliage (homoblastic) at the time of assessment. The ontogenetic change was shown to have an enormous impact on the composition of the dependent community, far exceeding the effect of genetic differences between the pure species and their hybrids. Dependent taxa exhibited a variety of responses to host hybridisation, however consistent with our other studies with eucalypts there was a clear trend for hybrids to support a greater number of dependent taxa than either pure species host. This study shows how the effects of genetic and ontogenetic variation in a forest dominant may extend beyond the individual and population to affect biodiversity at the community level. Our findings also argue that plant ontogeny can be a major mechanism driving community structure, which is deserving of much greater research emphasis in the study of biodiversity.

The importance of type (logs, snags or the top of trees) and treatment (shading or burning) of dead wood for its attractiveness to saproxylic beetles.

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Intensive forest management resulting in reduced fire frequency, the dramatic decrease of dead wood and changes in tree species composition in boreal forests is probably the main reason for the dramatic decrease of many saproxylic species in Fennoscandian boreal forest. This has lead to changes in forestry practise, including dead wood retention, which value not yet has been rigorously evaluated. To evaluate the value (from a conservation perspective) of different types of dead wood (logs, snag or remaining top of the tree) and find ways of improvement (burning and shading of logs) we performed a large-scale field experiment in northern Sweden. Species richness and the abundance of saproxylic coleopterans were higher on logs than on snags and tops but the different types of dead wood also attracted different beetle communities. Some red-listed species as the endangered *Monochamus urussovi*, was most common on tops and also found on snags, but not on untreated logs. Treatment of logs also influenced their suitability for red-listed species. Eleven, seven, five and four red-listed species was found on burned, control, shade net and naturally shaded logs, respectively. The implication of these findings for conservation strategies is discussed.

Herbivory alters plant litterfall dynamics: contrasting "afterlife" effects due to genetic diversity in susceptibility to herbivory

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Arthropod herbivory can alter forest ecosystem processes by changing litterfall chemistry and dynamics. Specifically, herbivores in different systems have been shown to 1) accelerate leaf abscission and thus, stunt nutrient resorption, and 2) elicit secondary chemical defenses from plants, both effects which alter litter chemical quality. Litterfall is an important input of carbon and nutrients; thus, any factor that alters the quality of plant litter can profoundly alter the decomposition rate of that litter and the subsequent rate of nutrient release. Plant defensive chemicals, which persist in litterfall, are manifestations of biochemically-based allocation tradeoffs. The balance of these tradeoffs is in large part genetically determined; therefore, plants inherit defense strategies against herbivores to a great extent. Using two experimental test systems, we illustrate how the contrasting influences of herbivore-induced changes in plant litter dynamics can contribute differently to the long-term dynamics of nutrient cycling in two different types of ecosystems, coniferous and broadleaf deciduous forests. Within a species, and population, variation in ecosystem processes may arise from genetic variation as expressed in susceptibility and resistance to herbivory. Among different species, within a community, the net ecosystem effect of herbivory may depend largely on plant phylogeny.

Estimation of conservation areas in the light of historical biogeography

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A method for estimating protect areas for biological conservations is newly introduced in the light of historical biogeography. Biological conservation is achieved in part by establishing protect areas for particular organisms. So far ecology has greatly contributed to the establishment of such nature reserves. Generally, ecological research only deals with synchronic biological phenomena. Biological conservation, however, should be made referring to historical events through evolutionary transition. In this respect, historical biogeography will play an important role in conservation biology. The vicariance model of historical biogeography is utilized to explain the present distribution of organisms. Under this model the distribution of particular species is most likely established through the process of extinction and/or fragmentation. The dispersal vicariance analysis (DIVA) used in the historical biogeography is applicable to detect highly fragmented species by using parameters of habitat fragmentations in spatio-temporal scale. The analytical procedure of the method is illustrated in the case of marine mite genus *Isobactrus* (Acari: Halacaridae). As the result of the protect area analysis, the south western and middle eastern parts of the Pacific were nominated as the most desirable areas for biological conservation of the mites.

Diversity of *Aegialites* - an intertidal rock dwelling genus (Coleoptera: Salpingidae: Aegialitinae)

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Aegialites Mannerheim, 1853 is an intertidal rock dwelling genus of amphi-Pacific distribution, which comprises 17 species in Asia and 13 in America. Its range includes the coasts of California, British Columbia, and Alaska, inclusive of islands, and Honshu, Hokkaido and 17 islands in the Far East of Russia. The range of *Aegialites* is largely congruent with that of north Pacific auks (Alcidae), also with that of seal species (Pinnipedia). The wingless *Aegialites* species are highly adapted to their special habitat by morphological, ethological, and presumably also physiological characters. The restricted range of the genus, the adaption to a cold climate (subarctic, cold currents) and an amphipolar vicariance with its sister group *Antarcticodomus* (subantarctic islands, Chatham Islands) imply a high phylogenetic age. Apparently, all the species are local endemics; non-adaptive radiation is hypothesized. 24 species, all from rocky seashores of the northern Pacific, are described as new. The genus requires further investigation: special physiological adaptions are still unknown and our current knowledge of the true diversity of the genus is far from complete.

The importance of conducting regional faunal surveys: a case study involving velvet ants in Wisconsin. (Hymenoptera: Mutillidae)

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Regional insect faunal surveys can be invaluable to taxonomic and ecological studies. Such surveys fill gaps in the existing distributional and biological knowledge for many taxa. Generally, trapping materials are inexpensive and trapping techniques are relatively low-effort, yet they often yield an abundance of new specimens for examination. Regional insect surveys are also critical for identifying and prioritizing areas targeted for conservation. Considering that insects typically constitute the majority of animal species for a given habitat, conducting surveys to identify which species are present is an important first step in any conservation effort. This case study illustrates the importance and value of regional insect surveys. Relatively simple, inexpensive survey techniques were utilized in Wisconsin to look for members of the family Mutillidae. Results of this study will include: 1) the publication of 25 new state species records (only four species had been reported prior to the study); 2) a sex association resulting in a new synonym for one species; and 3) a description of the previously unknown female of another species. Although this study focused on mutillids, the described survey techniques and materials are taxon-independent, and thus can be used by researchers anywhere who wish to study a variety of insects.

The arthropods of Morelos State: a preliminar synthesis

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Morelos, one of the smallest states in central Mexico (4 958.22 km^2), is located between two important biogeographic regions, neartic and neotropical. Climate conditions, physiography, and plant diversity make it an important zone in terms of species biodiversity. The purpose here was to compile information about all arthropod species recorded for the region. As many as 700 papers (from Mexican and foreign journals) were scanned. Up to date, 4,108 species are recorded included in 387 families and 40 orders within several classes: Arachnida, Acarida, Crustacea, Chilopoda, Entognatha, and Insecta. The Insecta grouped 3596 species (1509 of those were coleopterans). This information represents part of all the species potentially existing in Morelos. New investigations and revision of additional bibliographic material will surely increase the count.

Saproxylic beetle diversity & the cumulative ecological effects of wildfire & forest harvesting in boreal mixedwood forests.

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"Along with industrial forest harvesting, natural disturbances such as wildfire continue to play dominant roles in shaping the boreal forest landscape. Therefore, to achieve goals for biodiversity conservation and sustainable forest management, the individual and combined effects of natural and anthropogenic disturbances need to be better understood. In this study, we examined the effects of wildfire and forest harvesting on saproxylic beetle species diversity and composition to test the hypothesis that the combined effects of these two disturbances are greater than either disturbance alone. Specifically, we conducted a replicated, stand-level experiment in and around a large-scale (120,000ha) wildfire that occurred during the spring of 2001 in northern Alberta, Canada. Using flight-intercept traps, we compared saproxylic beetle assemblages from four stand treatments: (1) undisturbed; (2) burned; (3) clear-cut harvested; and (4) post-fire salvaged. Preliminary results suggest that fire and harvesting differentially affect saproxylic beetle assemblages and the combined effects of these two disturbances are cumulative for many (>20%) species. Moreover, our results demonstrate a link between the effects of disturbance, the breakdown of coarse woody material, and soil nutrient status, suggesting that alteration of saproxylic species diversity may have implications for soil nutrient dynamics in burned stands.
"

Mites as ecological indicators of sustainability in Michigan tree fruit

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An ecological indicator is a tool that can be used to detect disruption in the environment. In agricultural landscapes, it can be used to determine the relative sustainability of an ecosystem exposed to various combinations of management strategies. Mites are a rapid measuring tool of orchard health and its ability to cope with pest infestations, fertility, mulching, pruning, ground cover management, and many other orchard phenomena. Mites essentially integrate a wide range of management practices, environmental occurrences and surrounding habitat influences in and around the orchard. Monitoring is used to determine mite species presence and their relative abundance. These simple measurements, combined with seasonal ratings, are plugged into a mite index model. The mite index value is used to characterize the orchard along a continuum of orchard ecology classifications. An immediate goal of developing ecological indicators of sustainability is for tree fruit growers to use this information to make ecologically sound orchard management decisions. A more long term goal is for tree fruit growers to understand if their orchards are progressing positively toward a more sustainable and healthy system or negatively toward a disrupted system that will negatively effect yield, nutrient availability, pest populations, and non-target impacts.

Effects of conventional & integrated crop production on Collembola (Arthropoda: Hexapoda) communities in the soil of papaya orchards in Espírito Santo, Brazil

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Collembola have significant influences on soil microorganisms and fertility but information on these ecologically important arthropods is lacking, especially in agricultural and tropical environments. This research was conducted to identify species present and determine effects of agricultural practices on Collembola in Espírito Santo, Brazil. Soil samples (4.8cm diameter, 10cm deep) were collected in Feb, May, Aug and Nov 2003 from papaya orchards under conventional (traditional management and pesticide application) and integrated production (selective pesticide application only when necessary based on pest monitoring) in one experimental site with 3 treatments (conventional, integrated with drip irrigation, integrated with microsprinkler irrigation) and 4 commercial orchards (2 with conventional, 2 with integrated production). Berlese-Tullgren funnels were used to extract Collembola from samples. In initial results (February-August) collembolan density ranged from 13,000-187,000 Collembola/m^2. In the experimental site, *Willemia brevispina*, *Arlea* sp., *Folsomina* aff. *onychiurina*, *Folsomides* aff. *centralis*, *Folsomides* aff. *parvulus*, and *Cyphoderus* aff. *similis*, were more abundant with integrated versus conventional production. However, this difference was not observed in commercial orchards sampled. Results indicate that relatively high populations and diverse communities of Collembola occur in papaya orchards in Espírito Santo. However, no collembolan species were apparent as consistant biological indicators of conventional or integrated production.

Biogeography of butterflies (Lepidoptera: Papilionoidea) of the east slope of the Colombian Cordillera Oriental: their use in definition of priority conservation areas

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The study was made to propose priority conservation areas in the east slope of the Cordillera Oriental, based on a biogeographical analysis of butterflies (Papilionoidea); records from biological collections and literature were gathered to that end. Presence-absence matrixes were made using records and 341 quadrants of 7′ 30″ x 7′ 30″ resulting from the division of the study area. Cluster analysis and parsimony analysis of endemicity analysis were applied to them, and biogeographic units were defined and evaluated according to priority criteria for conservation. 8145 records of 1269 species were obtained, belonging to 62 municipalities of 10 departments that comprise the east slope of the Cordillera Oriental. Records were found only in 83 quadrants where 1098 butterfly species with specific geographic information were registered. The butterflies of the east slope these follow a distributional pattern determined by the influence of the transinterandean and cisandean biogeographical regions, differentiating two areas: the middle and south parts of the east slope, besides of being segregated by an altitudinal gradient. These areas were defined too as coincident with humid refuges and centers of endemism. The regions that gather the municipality of Mocoa (Putumayo), Piamonte (Cauca) and Ipiales (Nariño) were to be considered of priority in conservation efforts.

Detecting canopy insect fauna in a tropical rain forest

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A comparative study of trap systems for flower visiting beetles was carried out at Pasoh Forest Reserve, West Malaysia, in the canopy of a lowland tropical rain forest. We compared a balloon system (traps hung by balloon), a hanging system (traps hung by rope from a branch of an emergent tree) and a tower system. Abundance of scarabaeid beetles caught by traps was highest in the balloon system and lowest in the hanging system. The species composition of Scarabaeidae caught by the balloon system was quite similar to the tower system but was relatively different from the hanging system. *Dasyvalgus dohlni* and *D. pyrrhopygus* were absent in catches from the hanging system. *D. dohlni* and *D. pyrrhopygus* were therefore distributed in a relatively higher part of the forest canopy. This suggests that the hanging system was not so effective in catching beetles acting in the upper canopy and that the balloon system was useful in collecting these beetles because we were able to elevate the trap above the canopy layer.

Are endemic Curculionoidea (Coleoptera) good bioindicators of high biodiversity areas in the Iberobalear region of the southwest Europe?

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Endemic species represent a way to quantify the richness, diversity and habitat conservation status of geographic areas. In this way, the Ibero-Balear region occupies an outstanding position in Southwest Europe as it has a high concentration of endemic species and is considered as the origin point of a great number of other species that actually have a wider distribution. The results showed in previous works, allowed us to elaborate the Book of the endemic Curculionoidea from the Iberian Peninsula, Balearic and Canary Islands. In the Iberobalear region over 2000 curculionoidea species are known, from which more than 25% have been recorded as endemic of this region In the present work we have analized the distribution of these endemic species in the Iberian Peninsula and Balearic Islands, and we have compared it with the distribution of other arthropod groups, amphibians, reptiles and monocotyledonous plants. Although the studied groups differ in the levels of endemicity and in the width of the distribution ranges, almost in all of them distribution patterns of high endemicity areas are similar. There is a significant correlation between most groups and we also discuss if the endemic Curculionoidea are good bioindicators of high diversity areas.

Distribution map of genus *Exapion* (Coleoptera, Curculionoidea, Apionidae) in Spain

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Genus *Exapion* includes nearly 30 palaearctic species, although today its range of distribution has been increased due to the introduction of certain species (*E. fuscirostre* (F.) and *E. ulicis* (Forst.)) in other parts of the world as Australia, New Zealand and Northamerica. These introduced species have been largely used as control agents of also introduced weeds as *Ulex europaeus* L. and *Cytisus scoparius* (L.) Link. In Spain, is represented by 15 species that belong to two subgenus: *Exapion* with 10 described species and 4 subspecies, and *Ulapion* with 5 described species. Almost half part of these, are Spanish endemic species (six species), some of them restricted to small areas as *E. oblongulum* (Desbr.), *E. wagnerianum* (Schatzm.), *E. breviusculum* (Rosenh.) and *E. laufferi* (Schils.). The *Exapion* species more widely distributed is *E. fuscirostre* known from great part of the Spanish mountain range, due to its association with certain Genisteae species that belong to the genus *Cytisus* (*C. scoparius, C. multiflorus* (L'Hér.) Sweet, *C. oromediterraneus* R. Mart.) that are widely spread in the mountains of Spain. Distribution maps show that the Spanish province that have more species of this genus is Leon (in the north), with most of the records associated to the mountain systems (Montes de Leon and Cordillera Cantábrica)

Do summertime temperatures form hotspots of jumping spider endemicity?

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Previous sporadic and opportunistic collecting of jumping spiders (Salticidae) indicated that the southern and central-western regions of Australia are centres of endemicity. Recent broad-scale biogeographic surveys in Australia have revealed many new undescribed forms of jumping spider, indicating the presence of a very rich and diverse fauna. A broad-scale biogeographic survey of the jumping spider fauna (Salticidae) of the Western Australian Wheatbelt was carried out to determine the levels of diversity and their relationships with environmental gradients on a regional scale. A total of 41 genera and 121 species, of which 17 genera and 106 species are undescribed, were collected. Patterns in the species composition of assemblages across the survey area were broadly correlated with environmental gradients, particularly 'Maximum Temperature in the Warmest Period'. High proportions of apparently localised endemic species dominated the geographical patterns in species composition. Speciation in these 'hotspots' may be driving broad-scale biogeographic patterns in this area, but more detailed taxonomic and ecological studies are required to test this hypothesis.

Saproxylic invertebrates & wood decay fungi in living trees in Tasmania's southern forests

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Mature trees are an important part of a forest ecosystem providing a range of unique habitats that are utilised by both vertebrates and invertebrates. In the southern hemisphere little is known about the saproxylic organisms associated with living trees. However, in Europe many species of invertebrates and wood decay fungi associated with old trees are threatened, due to a severe decline in habitat availability. A long-term impact of current native forest management in Tasmania's wet eucalypt forests will be to alter stand age structure across the managed forest landscape, resulting in the decline of old-growth features. The effect this will have on the saproxylic communities is unknown due to a lack of knowledge of both the species involved and their habitat requirements. This poster describes two ongoing interrelated ecological studies of saproxylic communities that should put ecologists in a better position to advise on ways of sustaining the full spectrum of biodiversity in managed wet eucalypt forests. One is an investigation of the invertebrate assemblages found in living *Eucalyptus obliqua* trees of different ages (young regrowth to oldgrowth), and the other explores the relationships between fungal communities, stages of stem decay and increasing stem age.

Bear necessities: myrmecophagy of grizzly bears in north-central British Columbia.

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The grizzly bear (*Ursus arctos*) is a species of special concern in central British Columbia. Little is known regarding grizzly bear ecology in the Sub-boreal spruce biogeoclimatic zone, so a multi-year study was conducted between 1998 and 2002. As part of the larger study, we investigated bear myrmecophagy by direct and indirect assessment. Up to 62 radio-collared grizzly bears were located twice per week. Randomly selected bear locations were visited within 1-14 days, and a habitat-plot survey was conducted. Foraging was the primary activity identified at 385 of the 538 sites visited. Of 9 food-related activity categories, foraging for ants was the 4th most common. Myrmecophagy was infrequent in spring, but relatively significant in summer and fall.

Grizzly bears appeared to forage opportunistically for ants, and showed little preference for any particular species. Half of the bear scats with ants had a high proportion of ant remains of many species, indicating targeted "anting". The most prevalent species eaten were *Camponotus herculeanus*, *Formica neorufibarbis*, *Formica aserva* and *Lasius pallitarsis*, all of which are very common in the study area. This study shows that ants are an important food source for grizzly bears. Promoting ant habitat is recommended for grizzly bear management.

The rediscovery and captive breeding of the "extinct" Lord Howe Island stick insect, *Dryococelus australis* (Phasmatidae)

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Dryococelus australis was a large, impressive stick insect, abundant in the forests of Lord Howe Island until it was exterminated following the introduction of rats in 1918. In February 2001 three live specimens of *D. australis* were discovered feeding on the tea-tree, *Melaleuca howeana*, on Ball's Pyramid, a desolate, volcanic spire, 25 km SE of Lord Howe Island. Although about 30 bushes of *M. howeana* and other low vegetation were growing on Ball's Pyramid, the stick insect was found on only one *M. howeana* bush that was growing below a small water seepage. When Ball's Pyramid was visited again in 2003, 17 individuals were counted and two pairs were removed for captive breeding in Sydney and Melbourne. Both pairs have successfully bred, resulting to date, in over 200 eggs that have so far yielded 12 nymphs in late instar and two adults.

A comparison of insect assemblages associated with macrofungi in native & exotic New Zealand woodlands

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The fruiting bodies of macrofungi attract a specialist guild of fungivorous arthropods, along with their associated parasitoids and predators. The aim of this study was to compare the Coleoptera and Diptera found on decomposing fungi in four classes of New Zealand woodlands: native beech, native mixed deciduous, restoration/conservation sites and exotic plantations. Seventeen woodlands across the South Island were examined and, to standardize sampling, insects were collected using traps baited with commercially produced *Agaricus* mushrooms. In total, 9 families and 36 species of Coleoptera were collected, along with 14 families and 47 species of Diptera. As expected, the native beech and deciduous woodlands had the most diverse coleopteran faunas, with the restoration/conservation sites having the poorest. However, Diptera where most diverse in the exotic conifer plantations, with the mixed deciduous woodlands having the least rich faunas. The results indicate that conclusions regarding the diversity of woodland faunas can be dependent upon which taxa or functional guilds are considered. The results also suggest that monoculture plantations of exotic tree species need not necessarily contain less diverse faunas than native woodlands. The use of a 'site score' based on this specialist guild may have potential use as an indicator of conservation success.

Change in a butterfly community with forest growth in a cool temperate region of central Japan

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Transect counts of butterflies were conducted in the northern part of Ibaraki, central Japan at 22 census sites, composed of successive stages of forest growth (natural deciduous forest series and coniferous plantation series). The species richness, abundance and diversity indices of butterflies were high in the early stages of forest growth in both the natural forest and plantation series. In the natural forest series, typical natural forest species increased with the progress of secondary succession, and the old secondary forests and old-growth natural forests had similar species composition. Almost no typical natural grassland species were recorded in the young secondary forests. In the cutover land, the number of individuals of grassland species considerably decreased from the first to the second year after clear cutting. Thus, the suitable stage for grassland butterfly species lasts for only 1–2 years after clear-cutting because the regrowth closes over the ground vegetation after 3-4 years. Old secondary forests may be able to give refuge to the forest butterfly species, including typical natural forest species. In the plantation series, only a few typical natural forest species were recorded in the older stages, but the species richness and abundance of grassland species (incl. typical natural grassland species) were very high in the young plantations (up to 7th-8th year) because of frequent clearings by humans.

Effects of volcanic ash on insects in a tropical forest canopy

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The effects of volcanic ash on insects have been proven repeatedly in the laboratory, but field studies are few, and have utilized temperate-zone ground-dwelling species or colonial Hymenoptera. Our research focuses on an entire arthropod community in the tropical forest canopy, and at a still-intact habitat affected by volcanic activity rather than a devastated blast zone requiring recolonization. We investigated the impact of ash deposition levels on canopy arthropods from the West Indian island of Montserrat, which has been subject to ongoing volcanic eruption since 1995. Arthropod samples were obtained using canopy fogging at four sites regularly receiving different levels of ash deposition, and foliar ash was measured during each sampling. We compared our findings from Montserrat with samples from the neighboring island of St. Kitts, which has not been subject to recent volcanic activity. Based on analysis of 44,290 specimens in 34 samples from 4 sites on Montserrat and 3684 specimens in 3 samples from 3 sites on St. Kitts, volcanic ash appears to have a limited impact on the forest canopy insects of Montserrat. We explore potential environmental and evolutionary implications for these results.

Genetic diversity & phylogeography of endemic beetles on oceanic islands: examples from the Canary archipelago

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The Canary Islands constitute one of the 25 "hot spots" of worldwide biodiversity. Seven main volcanic islands, 110 km northwest Africa, form the island belt. They have a large diversity of habitats, including a low altitude xeric scrub, cloud and pine forests plus high altitude alpine areas. A fifth of the about 7000 species of the terrestrial invertebrates in the archipelago, are endemic Coleoptera. The effect of habitat fragmentation on the genetic variation levels of Canaries beetles varies among species with different life history traits and time since fragmentation. We focussed into two genera with quite different life histories; the endemic species of the genus *Pimelia* (Coleoptera, Tenebrionidae), present in arid and semiarid regions of all islands, and the carabid species-pair *Eutrichopus gonzalezi* and *E. canariensis*, occurring in the cloud laurel forest of the island of Tenerife. We have obtained COII mitochondrial sequences of *Pimelia* taxa and populations from several islands, and in *E. canariensis* and *E. gonzalezi* to study phylogeographic and demographic patterns. There are clear associations between volcanic events and genetic differentiation within and between taxa. The aim is to provide useful information helping in conservation management of the beetle populations and the island habitats they inhabit.

Diversity, biogeography and ecology of the insects of Australian rainforests– an overview

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Australian rainforests have been the subject of numerous studies since the late 1950s, mostly devoted to defining 'rainforest', mapping the distribution of rainforests and classifying them, based on structural and physiognomic features, and climatological parameters. The insects of Australian rainforests have only come into focus more recently, and the available information on them is reviewed in this study. Although rainforest covers only 0.3 % of Australia´s land area, it is particularly biologically diverse. Maps depicting the geographic distribution of Odonata, Cicindelinae, Rhopalocera and Formicidae show that these taxa are most species-rich in high rainfall areas, particularly those in the tropics. This distribution pattern, however, does not apply to all insects, Isoptera and Tettigoniinae being examples of taxa more species-rich in drier areas. Australian rainforests are well known for their evolutionarily conservative plants but are also home to several families of 'ancient' insects, such as the Peloridiidae, Xylophagidae, Micropterigidae and Agathiphagidae. Insects play many roles in rainforest ecosystems, as herbivores, decomposers, pollinators, seed dispersers, predator or prey. Rainforest insects represent textbook examples for camouflage and mimicry, but for many species it is still unknown how restricted their habitat requirements, and how host specific they are – information needed to protect their diversity.

Biogeography of the Australasian genus *Melobasis* (Coleoptera:Buprestidae)

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Melobasis is a primarily Australasian genus with around 200 known species in Australia, and around 40 known species from Papua. Within Australia the distribution patterns of species and subspecies suggest that speciation has been primarily the result of allopatric speciation at times of increased aridity. Only a few widespread species occur in the regions having less than 400mm of rainfall a year. A number of species from the less arid areas of S.W and S.E. Australia exhibit geographical variation, with the subspecies having distributions suggestive of differentiation in less arid refugial areas during drier periods, and in some cases more recent geographical overlap and introgression. The S.W. of Australia is the region of highest species richness. Within this region the transitional rainfall zone is much richer in species than the high rainfall zone and most of the endemic species occur in the former zone. Humid and sub-humid areas of S.E. Australia to S. Queensland, sub-humid central Queensland and the rainforest areas of Queensland south of Cape York Peninsula are also areas with many endemic species.

Discriminant analysis of the disruptive impacts of a horticultural mineral oil and two synthetic pesticides on citrus ecosystems

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The disruptive impacts of an *n*C24 horticultural mineral oil (HMO) and two synthetic insecticides (carbaryl and methidathion) on citrus ecosystems were evaluated in Peats Ridge and Kulnura on the central coast of New South Wales, Australia. At Peats Ridge, the impact of HMO was compared with carbaryl and methidathion in single trees, while at Kulnura, HMO was compared with methidathion in blocks of trees. Discriminant analysis was used to distinguish between the impacts of the different pesticide applications. Parameters representing scale pests and their natural enemies, as well as soil fauna, were included in the analyses. The model clearly distinguished the treatments into groups with a greater difference between the synthetic pesticide groups and the unsprayed control than between the HMO treatment and the unsprayed control. Results indicated that the HMO only had a minor and transient impact on the citrus ecosystems, while carbaryl and methidathion had significant and long-term disruptive impacts on the arthropod communities within these ecosystems.

Geographic variation in the water-based defence system of plants in the genus *Adriana*.

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Adriana (Euphorbiaceae) is an endemic genus of dioecious shrubs widely distributed in Australia. Extra-floral nectaries (EFNs) occur at the base of the leaves in the stipular position and are frequently visited by several species of ants. A survey of geographic variation in the size of EFNs of adrianas was conducted. Nectary sizes on plants collected from sites across Australia were matched with climate data from the nearest climate station. Data from three plant taxa were analyzed using multiple regression to relate gland widths to a series of climatic variables. Stipular gland widths exhibit significant geographic variation which is correlated with climatic factors, particularly the amount and variability of rainfall. Functional EFNs are found on plants of all three taxa and EFNs tend to be smaller on plants from areas of low rainfall. A droughting experiment was conducted in the glasshouse to examine the effects of water stress on extra-floral nectaries and nectar production. Numbers of active or functional nectaries on water-stressed plants did not decline more rapidly than numbers on watered plants. Reasons why water-based defences may still be effective for plants in arid regions are discussed.

Tube-renting wasps and bees monitored with nesting traps in Japanese forests

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"Nesting traps, which are typically made of drilled wooden blocks, reeds, or bamboo shoots, have widely been used to monitor hunting wasps or bees. This is because many of those species preferentially or exclusively nest in pre-existing tubelike structures, and because the nesting trap is easy to apply to various environments. We studied the tube-nesting hymenoptera with the nesting trap in cool-temperate forests of varying ages in Japan (36°N, 140°E) in order to learn how their species composition differ with forest conditions. A single trap was composed of 20 bamboo shoots and reeds, 4 to 16 mm in diameter, which were fastened together. We installed nine traps in each of six secondary deciduous broad-leaved stands, varying from 2 to >100 yr in age after clear-cutting, and in two conifer plantations, 5 and 70 yr-old. About 30 aculeate species utilized the traps in the 8 stands. The number of species recorded and the proportion of traps utilized were larger in younger than older stands both in broad-leaved and conifer stands. Spider wasps (Pompilidae) remarkably occurred in every stand in contrast with mason wasps (Eumenidae) or bees which only nested in young stands.
"

Macroinvertebrate diversity of isolated wetlands: are wetland quality and size important factors?

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Recent attention has focused on the conservation of isolated wetlands due to changes in the federal wetland regulatory program. In order to protect these isolated wetlands, more scientific information is needed to understand the ecology and function of these ecosystems. Our research focused on the role of different biological communities within isolated wetland ecosystems and how these communities change with environmental stressors (e.g., human disturbance). Specifically, the objective of this study was to compare macroinvertebrate diversity among isolated wetlands of variable quality (impacted or non-impacted) and size (large or small). Four depressional marshes were sampled from each possible combination of treatments (large impacted, large non-impacted, small impacted, and small non-impacted) from the Muskegon River watershed, Michigan, USA, during July 2002. Preliminary results suggest differences in macroinvertebrate diversity among wetlands of variable quality; however differences due to wetland size were not as defined. Knowledge of the association between macroinvertebrates and environmental stressors can be utilized in the assessment of wetland health, predicting change in wetland structure, and in the establishment of biocriteria for the regulation and protection of isolated wetland ecosystems.

Evolution of behavioural & ecological diversity: Australian *Acacia* thrips as model organisms

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A single lineage of phytophagous thrips has radiated on Australian *Acacia*, resulting in over 250 species in 30 genera. This radiation has generated four ecological suites of species: gall-inducing species (some of which have defensive 'soldier' castes), species that create a domicile by glueing together two or more phyllodes, parasitic species of these two, and opportunistic species that use old domiciles or other microhabitats. This study integrates ecology, behaviour, molecular phylogenetics and taxonomy, including the description of 140 new species. The driving force behind the system is the arid and unpredictable Australian climate that leads to intense competition for small protected spaces. These ecological pressures have generated a positive feedback mechanism, such that adoption and modification of host-plants by some thrips species create further niches for additional species. The causes and consequences of this behavioral-ecological diversification are explored through a novel, 'model clades', approach that provides insights into the evolution of social behaviour, host-plant use, and exploitative behaviours. The remarkable morphological, behavioural and ecological variation exhibited by these insects means that they can be considered a microcosm for understanding the processes that generate biodiversity among all phytophagous insects, and indeed among all animals.

Temporal and spatial variation of arboreal beetles diversity in an Amazonian rain forest

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Tropical communities are rich in diversity; however, many species have very local or patchy distributions, so that a complete understanding of species diversity requires comparison of species turnover at local scales. We assess the patterns of coleopteran diversity on different spatial and temporal scales at two 10 ha plots in western Amazonian forests. At each plot, we sampled diversity of seven groups of arboreal beetles by fumigation at 100 stations. During the sampling period, there was significant temporal variation in species richness due to the effects of El Niño Southern Oscillation and significant spatial variation amongst stations influenced by the architecture of the forest canopy within the two plots. Spatial autocorrelation analysis revealed that the richness and composition of this coleopteran assemblage was significantly patterned at distances below 1000 m for herbivores, fungivores and predators beetles, while dung beetles do not display any spatial structure. Such variation can strongly affect overall patterns of diversity and contribute to the high diversity characteristic of tropical coleopteran communities. Knowing the temporal and spatial patterns of distribution may be a fundamental step toward understanding basic patterns of species richness, hence achieving knowledge crucial to the evaluation of conservation effectiveness among conservation strategies.

Conservation of Galapagos Lepidoptera: a preliminary analysis of the endangerment status and factors affecting the Galapagos endemic Lepidoptera

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The Galapagos Lepidoptera fauna is characterized by poor diversification, high level of endemism and prolonged geographic isolation. These characters, because of their scientific importance, constitute unique attributes that emphasize, as a priority, their study and conservation. These unique attributes have been the foci of our research and conservation efforts for 15 years. So far, 310 species of Lepidoptera are known to occur on the archipelago and 52 % of the native component of this fauna is endemic. This study investigates the factors affecting the conservation of the endemic species and presents preliminary data on their endangerment status. The introduction of exotic species and the transformation of native forests into agricultural or urban areas are the main factors affecting the conservation of the endemic species.

The effects of human impacts on cave arthropod biodiversity: Thailand component

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Cave ecosystems are widely regarded as being one of the most vulnerable ecosystems on earth. They are remarkably fragile and subject to intense pressure from human activities. One of the most prevalent of these activities is the human utilization of caves for recreation and tourism. These activities are detrimental to the cave ecosystem is based largely upon the collective results of numerous, piecemeal observations. These observations have shown clearly that specific events and situations caused by people have an effect on the occurrence, absence or distribution of certain species. However, whilst useful at micro-management levels, this evidence can only be regarded as circumstantial when trying to establish links between cave use and the ecosystem as a whole, as required in macro-management. This study was carried out by conducting a replicated, natural experiment examining the overall effects of cave visitation and disturbance on the ecosystems of seven stream caves in southern Thailand, Meso and macro terrestrial arthropods were sampled in January, March and May 2003. A total of 4079 specimens were collected representing 519 species from 24 orders in 5 classes, showing the effects of disturbed areas having greater abundance than undisturbed areas and the highest diversity occurs near the entrance.

Geographical structure of morphological characters and mitochondrial DNA in *Semanotus japonicus* (Coleoptera: Cerambycidae) in Japan

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The sugi bark borer, *Semanotus japonicus*, occurs naturally in Honshu, Shikoku, Sado Is., and Oki Is. in Japan, and its main host tree is the Japanese cedar, *Cryptomeria japonica*. After intensive establishment of cedar plantations occurred during the 1960-70's, this borer has become a serious forest pest. To assess the geographical structure of *S. japonicus* populations, I used morphological and molecular information. Multivariate analyses were performed based on external morphological characters of the borer in Honshu and Shikoku. Comparing populations along the Pacific coast and populations by Wakasa Bay (on the Japan Sea), the reverse morphological features were indicated. Populations in the northeast and the southwest Honshu showed intermediate conditions. The patterns of morphological variations in *S. japonicus* are approximate to the distribution of the refugia of *C. japonica* throughout the glacial maximum. It is suggested that the geographical structure of the present *S. japonicus* population was caused by isolation during the last glacial period. Then I used partial sequences from mitochondrial DNA genes, the COI, tRNA$^{\text{Leu-UUR}}$ and COII to elucidate processes in the geographical structuring of *S. japonicus* populations. Two distinct lineages were detected, which probably represented the Pacific coastal and the Japan Sea side lineages.

Biodiversity of scarab beetles in Himalayan region of India & its correlation with weather parameters

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Scarab beetles, commonly called as White grubs, are the major limiting factor in agricultural crop production particularly during the months of April to October in the Himalayan region of India. Upland rice, finger millet, barnyard millet, French bean, black gram, chilies, tomato, brinjal, ladies finger, apple, pear, plum, pomegranate and forest trees such as *Quercus* sp., *Grevia optiva*, *Toona ciliata*, *Alnus nepalensis*, *Celtis australis* are severely damaged by the grubs. Due to changes in cropping pattern and profuse deforestation coupled with reduction in the population of predatory birds and other natural enemies, the white grub problem became alarming in the region. More than 40 species of white grubs belonging to subfamilies viz., Melolonthinae, Rutelinae, Dynastinae, Cetoniinae, Troginae, Scarabinae and Aphodinae of family Scarabidae have been recorded from the region. Attempt has been made to correlate white grub beetle population with the climatic factors *viz.,* maximum temperature, minimum temperature, relative humidity and rainfall. Data of light trap catches of white grub beetles of 13 years revealed significant correlation of weather parameters with beetle emergence. The present paper discusses species diversity and critically analyses impact of climatic factors on beetle population in the region.

Biology and distribution of the sugarcane planthopper genus *Eumetopina* (Hemiptera: Auchenorrhyncha: Delphacidae)

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Sugarcane has a very interesting Auchenorrhyncha (Hemiptera) (leafhopper and planthopper) fauna and among them the planthopper genera *Perkinsiella* and *Eumetopina* are of special significance. *Eumetopina* species are confined to SE Asia (although two African species are currently placed in the genus) and distributed from the pacific islands to Vietnam, but the greatest number of species seem confined to Papua New Guinea, the origin of commercial sugarcane. *Eumetopina* species live exclusively on sugarcane and are found in the growing shoot of the cane. When found they may be very common (hundreds per plant) and are frequently attended by ants of various genera. Around nine species have been described from SE Asia- mostly from Papua New Guinea but perhaps another ten species remain undescribed. One species, *E. flavipes* Muir is known to spread a phytoplasma disease of sugarcane (ramu stunt) in PNG and is found quite widely distributed on some Pacific islands. The biology, taxonomy and distribution of the known and undescribed species will be reviewed.

What wasp is that ? An interactive key to the identification of Australian & New Zealand ants, bees, sawflies & wasps.

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The Hymenoptera is one of the largest terrestrial invertebrate groups and can be found in most habitats including intertidal and freshwater environments. They perform important functional roles in natural and agricultural communities as herbivores, pollinators, and as predators and parasitoids. This interactive key provides an easier alternative to traditional dichotomous keys for enthusiasts and professionals to identify families of Hymenoptera occurring in Australia and New Zealand. The CD also provides comprehensive information about the order. The morphological characteristics and the biological traits for each family are well illustrated and clearly explained. In addition, a review of past and present phylogenetic classifications of the Hymenoptera is concisely laid out. A thorough bibliography is also included which allows users to easily access the major titles in the study of the Hymenoptera.

The cicadas of central eastern Australia

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This website is designed as an interactive identification guide to the cicadas of central eastern Australia. It covers an area extending from the Wet Tropics in northern Queensland south to the south coast of New South Wales and west to approx. 144°E. General information is provided on cicada endemicity, feeding, life histories, common names, biology, phenology, sound production and reception mechanisms, sexual communication, calling songs, habitats, defining species limits, species of economic importance, collection and literature. Specific information is given for the two families, Tettigarctidae and Cicadidae, 33 genera and 230 species within the region. Text detailing the appearance, size, distributions, habits and songs is provided for all species illustrated, along with photos, distribution maps and sound recordings, where available.

The distribution & conservation status of gall-forming midges from acacias in southern Australia

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Acacias of southern Australia support a species-rich fauna of gall-forming Cecidomyiidae that utilise inflorescences for their development. Most taxa are undescribed but can be identified using insect morphology, DNA sequences, gall morphology and biological characteristics. Eight broad groups of Australian midges were recognised based on gall and life history characteristics that encompass 22 species of *Dasineura* and eight species of *Asphondylia*. The conservation status of gall-forming cecidomyiids of *Acacia* was determined by scoring four host related factors (abundance, reservation status, host-organ availability, horticultural utility) and five insect related factors (abundance, specificity, temporal constraints, spatial constraints, reproductive capacity). Using insect-related factors alone, eight cecidomyiids had low score values, and when combined with host-related factors, four species had low total scores indicating potential for conservation concern. Surveys in 400 km grid squares indicate eastern Australia has a higher species richness of gall-forming cecidomyiids than Western Australia with most species (16) recorded from the northern New South Wales-southern Queensland region. There was a strong segregation in the faunal distribution between eastern and western Australia, with only two species shared between the two regions. Two eastern Australia species, *D. rubiformis* and *D. pilifera* were found naturalised and abundant in Western Australia.

Himalaya: evolution of a "hot spot" area - insight from phylogenies of sericine chafers (Coleoptera: Scarabaeidae: Sericini)

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Due to extent in width and height, the Himalaya has an outstanding position within the tertiary orogenetic belt at the southern margin of the former Cretaceous Asian continent. In the present study phylogenies were generated for ten genera/species' groups of sericine chafers (Coleoptera: Scarabaeidae) with different patterns of vertical and horizontal distribution. The cladistic analyses were based on morphology, mainly characters of external skeleton and genitalia. These hypotheses of phylogeny provide evidence to predict multiple events of radiation which may be regarded as a main source of the high diversity within this region. The occurrence of numerous basal lineages in the Himalaya, additionally, lead to assume that the Himalaya was in historical framework a „refugium", too, being able to buffer fluctuations of climate by "orobiom-shifting". The phylogenies found show a congruent pattern in groups with similar vertical and/or horizontal distribution. Patterns of vertical and/or horizontal distribution are shown to be of crucial importance for gene flow, dispersal capacity, and for the opportunity to occupy new habitats. Hence, in framework of evolutionary process they should have influenced not only patterns of diversification but they determined recent faunal composition (obviously, beside the influence of recent climate) of the Himalaya, too.

Diversity of arthropod fauna associated with rice ecosystem in Nagaland, India

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Large numbers of diverse arthropods are associated as pests and beneficial with rice ecosystem in Nagaland. Rice is the staple food, which is traditionally cultivated under jhum, terrace and low wetland farming depending on the topographic conditions. For an effective pest management, identification of key pests and their natural enemies is very important. The present investigation was therefore, conducted during 2002 to 2003 under terrace farming located at Jotsoma area (1070 MSL) to find the qualitative and quantitative composition of arthropod faunas.Twenty-one families of seven orders were recorded as aerial arthropods with maximum number in Anisoptera and Zygoptera. Twentyfive families of eleven orders represented the nightactive arthropods where Muscidae followed by Blattidae and Meloidae were recorded maximum. The terrestrial arthropods were represented by eighteen families of ten orders with Collembolla and Formicidae being dominant. Few groups of Cryptostigmata, Collembola and immature Scarabaeidae were recorded from soil inhabiting arthropods with Collembola as dominant group. It also showed that terrestrial arthropods were most abundant followed by aerial arthropods with different group dominating different faunas. Besides, natural enemies such as *Cicindella sp. Coccinella septumpunctata, C. sexmaculata, C. transversalis, Agriocnemis pygmaea, A. femina femina, Tetragnatha sp., Oxyopes sp., Agriope sp,* and Theridid spiders were abundantly found.

Evolutionary radiations of ants in arid Australia

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Ants are undoubtedly one of the biggest biological success stories in Australia, and their story is played out primarily in the arid zone. Ant species richness, productivity and behavioural dominance are all far higher in arid Australia than in climatically comparable regions elsewhere in the world. Semi-arid southern and northern Australia support by far the richest communities of epigaeic ants known on Earth, with over 100 species commonly occurring within 0.2 ha. However, ant genus richness is only moderate, with its extraordinary species richness due to exceptional speciation within a relatively few genera. The most important genus ecologically is *Iridomyrmex*, whose species exert a level of behavioural dominance unparalleled elsewhere in the world outside the canopies of lowland tropical rainforest. The three richest genera, *Melophorus*, *Meranoplus* and *Monomorium*, each with huneds of species in arid Australia, possess remarkable adaptations that indicate a long evolutionary association with behaviourally dominant species, pointing to unusually high ant productivity over evolutionary time in arid Australia. A range of historical and ecological factors is likely to contribute to the remarkable ant diversity in arid Australia, but there is emerging evidence that carbohydrate supply might be the key driving force behind exceptional productivity and behavioural dominance.

Natural enemies & the evolution of resistance to Bt crops in herbivorous insects

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Some recent theoretical research has suggested that natural enemies can retard or accelerate the rate of evolution of resistance to Bt crops in an herbivorous insect. Other theoretical results imply that natural enemy induced mortality may have little influence on the rate of evolution. I provide an overview to the various theoretical approaches to the problem of resistance evolution and focus on the role of natural enemies in modifying evolutionary patterns. I suggest some conditions where natural enemies could exert considerable selection pressure on an herbivorous insect to adapt to Bt crops.

Indicators of climate change: insect herbivore colonisation of a novel host plant in a warmer climate

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Rapid climatic warming over the next few decades will affect the distribution and abundance of many species. We conducted a transplant experiment to investigate the potential colonisation of a plant species by insect herbivores under a warmer climate. Plants were transplanted to two sites, 280km north of the host-plants current coastal range; the transplant sites were 1.2 and 6.5^{0}C warmer than the northernmost and southernmost boundaries of the species' current range respectively. We compared the structure and composition of the herbivorous Hemiptera and Coleoptera communities on *Acacia falcata* transplants to that of *A. falcata* within its current distribution and to a closely-related *Acacia* species that naturally occurred at the transplant sites. Guild structure of this herbivore community did not significantly differ between the transplants and that found on *A. falcata* within its natural range, but did differ between the transplants and *A. leptostachya*. Host plant identity was apparently more important than climate in influencing the structure of the colonising herbivore community. If this result holds for other plant-herbivore systems, we might expect that under a warmer climate, broad patterns in community structure may remain similar to that at present, even though species composition may change substantially.

Endemic radiations within the megadiverse fauna of Australian Hymenoptera

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The Australian hymenopteran fauna is huge, poorly studied, and probably 5-10 times larger than the currently described 8400 species. Because of its long post-Gondwanan isolation, a large proportion of the fauna is endemic, although numerous groups share species with New Guinea and other islands to the north. Although detailed assessment of the level and degree of endemic radiations is hampered by a lack of supporting phylogenetic information, several groups are recognised for their remarkable explosive radiations into particular unique austral habitats or host groups in the case of parasitoids. Further, a number of little known, poorly studied taxa have been shown recently to have speciated far more in Australia than on any other continent. Here we use a number of examples, mostly from the parasitic Hymenoptera, to examine the extent, uniqueness and nature of these radiations in Australia. These examples come from the superfamilies Evanioidea, Platygastroidea, Ichneumonoidea and Chalcidoidea and will focus on parasitoids of native bees and wasps, Coleoptera, Lepidoptera, the spider fauna, and importantly 'parasitoid' groups associated with galls of *Eucalyptus*.

Relevance of edge & riparian effects to conservation of litter beetles in streamside reserves

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Streamside reserves in the form of unlogged strips of forest 20-30 m either side of streams, are a component of the reserve network within Tasmanian logging areas. Although primarily protecting aquatic values, these reserves have the potential to provide habitat and landscape connectivity for terrestrial litter-dwelling beetle species unsuited to conditions in coupes or young regeneration forest. Narrow reserve areas are prone to edge effects, which could limit their effectiveness for some species. Preliminary results indicate that edge effects on litter beetles usually extend for approximately 10 m or less from coupe edges in wet eucalypt forest. Litter beetles also respond to the riparian zone, with generally lower abundance and species richness and changing community composition for 5-10 m from streams. The combined effects of these riparian and edge zones may render streamside reserves unsuitable for some species. 100 m wide wildlife habitat strips appear to have greater potential than streamside reserves for protection of terrestrial invertebrate fauna.

The relationship between crown structural attributes & the canopy invertebrate fauna of 100 year-old & old-growth Tasmanian *Eucalyptus obliqua* trees

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Invertebrates were collected in the crowns of 100 year-old and old-growth Stringybark *Eucalyptus obliqua* to compare the composition of invertebrates associated with specific regions within younger and older tree crowns. The aims of the research are to test the hypotheses that: a) older trees are inhabited by a different invertebrate community than younger trees; and that b) older trees are quantitatively more structurally complex than younger trees. Rope techniques were used to access the crowns of 16 trees near the Huon River at Warra LTER in Southern Tasmania, Australia. Spatially explicit trapping methods (sticky traps, omnidirectional flight intercept traps, and bark crawl funnel traps) were selected to compare live branches, dead branches, and the trunk surfaces in the upper and lower crowns. Structural measurements on branching structure and special habitat location were used to record the exact position of the traps in the tree crown, and processed into 3-dimensional virtual tree models to aid other researchers and the general public in visualizing these arboreal habitats.

A plethora of ants, a paucity of ant pollination systems

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While the Hymenoptera are among the most important pollinators worldwide, the ants, although abundant in most terrestrial ecosystems, and frequent flower visitors, are rarely involved. I briefly review the reasons for the scarcity of ant pollination and the few cases in which ant pollination has been documented. Two very different kinds of ant pollination system have evolved. The first, limited to two highly specialized interactions involving complex floral traits and insect behaviour, occur only in y, grassy or shrubby habitats in southern and eastern Australia. By contrast, the second kind s relatively unspecialized, with easily accessible floral rewards that ants share with a variety of small winged or flightless insects. These systems occur chiefly in harsh habitats where selection for plants with low, accessible growth forms favour visits by ants. The discovery of more plant species with varying levels of dependence on ants for seed set seems likely. Moreover, the apparently unspecialized ant pollination systems may reveal behavioural, morphological, physiological, chemical or colony traits that predispose particular ant species for pollination.

Phylogeography of two genera of funnel web spider (*Haonyche* & *Atrax*) from the saproxylic habitat in Tallaganda State Forest (NSW)

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Decomposing logs are habitat to invertebrate species occurring across a spectrum of ecological niches. A collaborative research project is examining patterns of genetic endemism among saproxylic (log-dwelling) invertebrates across Tallaganda State Forest (NSW). Previous study of the onychophoran *Euperipatoides rowelli* revealed strong population structuring, identifying five areas of local endemism. Subsequently, *E. rowelli* has been used as a base-line study species for a comparative phylogeographic study of saproxylic invertebrates. To date, analysis of the mitochondrial gene Cytochrome Oxidase I (mtCOI) has revealed that a number of taxa display phylogeographic congruence with *E. rowelli*, suggesting that the saproxylic habitat has been a major factor in the evolution of these invertebrate species. *Haonyche* and *Atrax* are two ecologically similar genera of funnel web spider found across Tallaganda State Forest, however, *Haonyche* species are log-dwelling whereas *Atrax* species are ground-burrowing. The niche partitioning of unnamed species within these genera allows the separation of species and habitat as contributing factors to the patterns of local endemism found in this system. The resolution of this question is central to defining the impact of habitat on the evolution of log-dwelling invertebrates and critical to the establishment of conservation priorities for invertebrate species in Tallaganda State Forest.

Biogeography of the coffee berry borer *Hypothenemus hampei* **(Ferrari 1867; Coleoptera: Scolytidae) and design of molecular markers**

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Hypothenemus hampei, **the most destructive insect pest of coffee, reproduces as matrilineal lines ensuring low genetic variability. To examine the biogeography of this pest, AFLP was performed to generate DNA fingerprints from a worldwide collection. The greatest genetic variability was found in the presumed center of origin. Samples collected from West Africa and Asia were more closely related to samples collected from the Americas than to samples collected in East Africa. The greatest amount of variation in the Americas was discovered in Brazil, and the distribution of that variation suggested that these lines spread from Brazil throughout Latin America. The presence of all American lines in Colombia was used in order to understand the distribution of** *H. hampei* **throughout the Americas. Furthermore, this examination revealed the origin of** *H. hampei***'s latest introduction, as well as the possible pattern and manner in which this insect was distributed in the Americas. We found at least 3 introductions of** *H. hampei* **in Colombia; however, a main line was found in every coffee producing area. Hypothesis for this dispersion will be presented. The development of molecular markers and their use in population genetics, biology, and ecology studies will be discussed.**

Lepidopteran diversity hotspots in Canada: use of Biodiversity World virtual laboratory for predicting past & future patterns

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British Columbia (BC) has the most diverse Lepidopteran fauna in Canada. Its postglacial origin makes butterflies ideal candidates for time-slice bioclimatic modelling. If adequate and precise distribution data were available, it would be possible to model the Lepidopteran diversity at various times – from the arrival of this group in BC (*c.* 15,000 yr BP) to its future distribution under changing climate. Canadian specimen information network held by the Canadian Biodiversity Information Facility provides a Lepidoptera data set (*c.* 100,000 point records) from which to create bioclimatic envelopes of butterfly species. Climate Space Model, which uses past and future climate scenarios and a set of point observations to capture a model environment, is used for building bioclimatic envelopes for each of the species in the data set. Many such envelopes are superimposed to generate species richness maps in Worldmap (http://www.nhm.ac.uk/science/projects/worldmap). The Biodiversity World project is developing a problem solving environment (PSE) to bring all these components together in a GRID setting. As an exemplar study of the PSE, the species richness maps, generated based on the distributions of butterfly species, are used for prediction of past and future hotspots of Lepidopteran diversity and for their comparison with the present hotspots.

Biogeographic connections between Australia & New Caledonia

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Two genera of Dolichopodidae (Medeterinae), *Atlatlia* and gen. nov., are sister groups known only from Australia and New Caledonia. In Australia both display a classical "Bassian" distribution pattern (Tasmania, eastern Australia to SE Qld., and southwestern Australia), typically associated with Gondwanan taxa, while in New Caledonia, they occur above 500 m. These two genera have radiated on both landmasses, but are unknown from New Zealand, New Guinea, Solomon Islands and Vanuatu. These taxa are highly unlikely to have dispersed between Australia and New Caledonia. Possibly some of these genera will be found in montane New Guinea, where the Highlands often hold old Australian elements of Gondwanan affinity. However, they are unlikely to occur in the tropical Solomons and Vanuatu, and would not have reached New Caledonia via island "stepping-stones". The Australia and New Caledonia pattern suggests a relictual Gondwanan distribution. Were these genera once widely distributed in eastern Gondwana? Or did certain groups have Australia and New Caledonia as a center of origin? The absence of the two genera from New Zealand is more puzzling. However, during the Tertiary, New Zealand underwent major climactic and tectonically induced extinction events, losing much of its ancestral flora, and probably associated insects as well.

Insect diversity on tree trunks

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Tree trunks have a characteristic but little studied invertebrate fauna. Sticky traps (plastic strips coated with Tanglefoot) were placed on eucalypt trunks in forests of northeastern N.S.W., at 12 sites over four seasons. Approximately 103, 000 arthropods were collected, representing some 180 families. Diptera comprised 85% of the total catch and the top nine family ranks, Sciaridae being first at 49%, the most abundant family on all traps, all seasons. This data is discussed with respect to taxonomic richness, trap bias, seasonality, and site history. The importance of soil fauna, hidden larvae, and "orphan taxa" are discussed. Artificial sticky traps and naturally exuded tree resins are similar biotic "traps", and therefore taphonomic analogues. Relative taxonomic abundances from modern trunk sticky traps and early Tertiary Baltic Amber collections are briefly compared, which suggest similar arthropod communities are associated with tree trunks through time.

Ant assemblage structure in the CFR: Investigating patterns across altitude

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Climate-modeling exercises have demonstrated that the fynbos in the Cape Floristic Region is likely to be highly sensitive to climate change and it is thought that this vegetation type will be wholly replaced with a different, unknown vegetation type. It is thus crucial to collect baseline data on taxa that are critical for ecosystem functioning in this biome. Therefore, we investigated ant assemblage structure across an altitudinal gradient ranging from sea level (Lambert's Bay) to approximately 2000 m above sea level (Sneeukop, Cederberg) and down again to 500 m above sea level (Wupperthal) in the Western Cape, South Africa. Sampling was undertaken during October 2002 and March 2003 at 17 altitudinal bands, stretching over three vegetation types (Strandveld, Mountain Fynbos and Succulent Karoo). Ant assemblages differed significantly with regard to altitude, geology and vegetation type. Four ant species were specific to the Succulent Karoo biome, and one species each to Strandveld and Mountain fynbos. Furthermore, one ant species was specific to the summit. Mean daily temperature, available area and the proportion of sand in the soil were the most important environmental variables explaining assemblage structure. These results suggest that the ant assemblages might also be influenced by climate change.

Distribution of invasive/endemic cockroach species on two oceanic islands

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Establishment of invasive species is one of the major factors causing decline of native faunas. Their impact is more severe on oceanic islands where faunas are more sensitive to environmental changes and more prone to extinction. Our study was centred on two oceanic islands: Mayotte and La Réunion. We describe the distribution areas of nine invasive and four endemic cockroach species. Our aim was to evaluate the impact of invasive cockroach species on the distribution and maintenance of endemic species. Interference between invasive and endemic species was evaluated by comparing their distribution areas, their habitat selection and their co-occurrence. Habitats occupied by invasive species highlight their colonizing capacities and their wide tolerance with regard to environmental parameters. Their distributions overlap greatly and many sites are colonized by guilds. Their presence is related to low elevation, presence of water and strong anthropization. Thus, irrigated sugar-cane fields on La Réunion are the most suitable habitat for invasive species, harbouring guilds of up to seven species. On the other hand, distributions of endemic species never overlap, although two of them co-occurred with invasive species. Their distribution areas emphasize their reliance on their original habitat and the threat that anthropization represents for their conservation.

Role of planted forests as habitat for the endangered *Holcaspis brevicula* & other ground beetles

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The Canterbury Plains in the eastern South Island is one of the most modified regions of New Zealand with less than 2% of indigenous vegetation remaining. From 2000-2004 we surveyed ground beetles using pitfall traps in several remnants of the formerly extensive *Kunzea ericoides* shrubland, a 7000 ha plantation forest, grassland and pasture areas, exotic shrubland, and the nearest mountain beech forest in the foothills of the Southern Alps. A total of 5985 carabids representing 43 species were collected, including two specimens of the critically endangered *Holcaspis brevicula*, a local endemic thought to be threatened by habitat loss and fragmentation. Catches per trap were lowest in *Kunzea* shrubland and highest in pasture and young pine plantations both of which had a large proportion of introduced species. By contrast, virtually all carabids trapped in shrubland, beech forest and old pine plantations were indigenous. All known specimens of *H. brevicula* were caught in the plantation forest. The small and fragmented *Kunzea* shrubland remnants may not be sufficient to maintain populations of all forest carabid species. We propose that the pine plantation provides an important substitute habitat for forest carabids.

Improving management for the Eltham copper butterfly (*Paralucia pyrodiscus lucida*) in Victoria, Australia

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The Eltham Copper Butterfly *Paralucia pyrodiscus lucida* (Lepidoptera: Lycaenidae) was widely considered extinct until its rediscovery in 1987. Public campaigns promoted the establishment of reserves to protect known colonies, and since 1988 the relevant management authorities have been involved in maintaining viable habitat for the butterfly. *P.p.lucida* is a widespread but isolated sub-species, with remnant populations in suburban Melbourne, Castlemaine (central Victoria) and Kiata (North-West Victoria). The butterfly was one of the first insects listed as Threatened under the Flora and Fauna Guarantee Act. All known colonies are managed individually by their local management authorities. Contrasting attitudes both of management agencies and funding bodies have had both positive and negative outcomes by maintaining interest of independent agencies, but not allowing for holistic state-wide management of the butterfly. The Eltham copper is an important flagship species for conservation in Victoria. Thus an integrated management regime has the potential to provide a framework for other potentially threatened species in Australia. This paper highlights the weaknesses in current contrasting management schemes and offers tentative solutions to strengthen management through promoting more effective liaison and coordination between all agencies involved.

Biodiversity & biogeography of Lord Howe Island: an answer in the Pacific

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In the 1950s the dipterist Paramonov labelled Lord Howe Island as "a riddle in the Pacific". It has remained both an enigma and an attraction to entomologists because of its great species richness and endemicity. The Australian Museum has had a long scientific association with the island, with surveys dating back to the 1860s. Over the past four years we have undertaken an All Taxa Terrestrial Invertebrate Inventory, including a synthesis of historical information. Although the Lord Howe Island fauna is predictably disharmonic, there is considerable representation of higher taxa, with 28 orders, 306 families and 903 genera recorded. At present, about 1600 species have been recognised, with overall endemicity estimated at about 40%. This endemicity reflects considerable within-island diversification, which is partly correlated to habitat types and altitudinal gradients. The origins of the fauna of the island are complex in that numerous biogeographic area relationships are recognised for Lord Howe Island, including: i. Indo-Pacific; ii. Gondwanan; iii. temperate Australia; iv. tropical North-east Queensland; v. New Caledonia; and, vi. New Zealand. These patterns are partly explained by classical biogeographical theory, namely proximity to source areas. However, the area relationships are also a reflection of Lord Howe Island's refugial nature and its geological history.

Species richness & abundance of ant-like litter beetles (Staphylinidae: Pselaphinae) in three forest types of northeastern New South Wales

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The species richness and abundance of ant-like litter beetles is examined for the fauna of three forest types in northeastern New South Wales, with a total of 229 species taken. Sampling was conducted for over a year in both managed and old growth stands of dry sclerophyll, wet sclerophyll, and temperate rainforest in the Styx River State Forest. Biweekly samples were taken of rotten wood and leaf litter, and from two flight-intercept traps (FIT's) at each site. The FIT's produced over 90% of the species taken, and half of the specimens. Species overlap between FIT's and the faunas characteristic of leaf litter and rotten wood was low, and these traps are productive in sampling for those species living in habitats that were apparently overlooked. Old growth sites usually produced larger numbers of species and abundances, and often were not strongly marked. The rotten wood fauna is characterized by high diversity and low numbers, while abundance of the leaf litter fauna is much higher.

Endemic insects of Andaman & Nicobar Islands

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This is the first comprehensive study on endemic insects of Andaman and Nicobar Islands, studied by the author during the long stay of eight years in these islands. Andaman and Nicobar Islands comprised of an arcuate chain of more than 500 islands, islets and rocks spreading in more than 1100 km in Bay of Bengal. The long isolation of these islands from the sub-continent has resulted in high endemism particularly in terrestrial faunal elements. Due to the sea barrier, the distribution of fauna in these islands varies from island to island. Some species are restricted to certain areas in Andaman's, some to Nicobar, while others are common to both the group of islands. Sometimes, a species is even confined to a particular island. These islands with 0.25% of the geographical area of India harbor about 4.2% of all known species of insects in the country. Presently, 2506 species of 1552 genera under 20 orders of insects have been recorded, of which 604 (24%) are endemic to these islands. At species level endemism is being recorded highest, in Trichoptera (70.8 %) and least in Blattaria (6.6%) percent, while in sub-species level, more than 70% endemism is recorded among the butterflies.The study indicates that the endemism may be more if the species are studied up to subspecies level with the molecular techniques.

Pacific voyagers: the diversification & decline of *Rhyncogonus* (Curculionidae: Coleptera) in the Pacific

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Surprisingly, large, flightless weevils constitute a conspicuous element of many oceanic island faunas. The genus *Rhyncogonus*, for example, occurs across Polynesia, on some of the most remote islands in the world. Multiple *Rhyncogonus* species occur on many oceanic islands, often sympatrically, which could represent independent colonization events or *in situ* diversification, linked with geographic isolation, morphological changes and host shifts. Studying the phylogenetic relationships within this genus provides an interesting opportunity to investigate diversification and colonization processes. Recent collections of *Rhyncogonus* from French Polynesia are compared with historical collections made by the Pacific Entomology Survey, over 70 years ago, highlighting the general plight of endemic island biotas and the need for further survey work in this region. Phylogenetic analysis of 2,000 b.p of nuclear and mitochondrial sequence data, for over 60 ingroup species, and numerous outgroup taxa, reveal that *Rhyncogonus* has affinities in the western Pacific, and would have had to colonize the eastern Pacific against prevailing ocean and wind currents. The long branches and deep splits in the phylogeny reveal a long history for this group in the Pacific region, colonizing new islands long before the first Polynesian canoes, and probably before the emergence of some oceanic island chains.

Ant functional group responses to perennial shrub loss & recovery in a xeric environment

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Habitat recovery within chenopod shrublands in South Australia, as in most arid environments, is extremely slow. Arthropod communities may be able to provide an indication of small scale recovery in these habitats because of their rapid turnover and close ties with ecosystem function. In bluebush (*Maireana* sp.) dominated chenopod shrublands, ants are one of the most abundant and species rich arthropod groups. Although this group has been touted as the quintessential indicator of ecological change, the evidence for their applicability in arid environments is so far unconvincing. Ant functional group composition within wetter, forested habitats appears to be largely controlled by shade and litter cover. However within ier, more open environments where access to sunlight is not limited, factors such as access to food resources are likely to be of greater importance. In this study, patterns within the bluebush shrubland ant community are therefore correlated with environmental variables, and habitat manipulations of shade and structure are used to provide insight into the mechanisms iving functional group level responses. This project will develop a greater understanding of the role of ants as bioindicators in arid environments and provide land managers with suitable tools to monitor habitat recovery.

Evolutionary radiations: what they are & how we can test for them

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A radiation results when the rate of generation of new taxa is greater than the extinction rate. Thus, any lineage comprising more than one taxon has undergone a detectable radiation. Most interest is centred on radiations that contribute to our understanding of evolutionary processes such as speciation, adaptation and biogeographical change. Typically, these include radiations that are characterised by changes in the rate of speciation, including rapid increases associated with innovations, ecological shifts or climatic change, and radiations that generate or assess biogeographic hypotheses. An emerging feature of phylogenetic studies is that the rate of origin of species does not appear to be constant across all taxa or through time. Some lineages appear to have undergone radiations in which taxa diversified by rapid speciation, interspersed with periods of extinction or relatively slow diversification. Comparative methods and the inclusion of a time dimension in phylogenetic studies allow increased understanding of evolutionary radiations.

Estimating biodiversity: a case study on true bugs in Argentinian wetlands

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Measures of biodiversity provide the base-line information on distribution, richness, and relative abundance of taxa required for conservation decisions, studies of ecosystems ecology, cladistic biogeography, and phylogenetic measures of conservation value. Species richness in particular is an increasingly important statistic in conservation evaluation. This work aims to improve knowledge of the Ibera complex (Corrientes, Argentina), one of the most important but least known South American wetlands, and of terrestrial heteropteran biodiversity. We use quantitative measures of inventory completion and statistical analyses to critique our results and to elucidate the effects of inventory design parameters, with a view towards increasing the efficiency and effectiveness of biological inventory.

Seven collectors sampled the fauna of true bugs in the Iberá watershed (Col. Pellegrini, 28º 32´ S 57º 09´ W) during early spring, summer, and late summer of one year. Sixteen families, 225 spp. and 4,688 adults were found. The lower bound of annual heteropteran species richness at Pellegrini was about 250-300 species. Among heteropteran families, the particular inventory methodology was especially efficient in sampling Miridae.

Aquatic wood – an insect perspective

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Aquatic immersed wood (snags) provide major substrate for macroinvertebrates and refugia for fish, turtles and amphibians. The effects of human intervention such as wood removal for navigation and angling on fluvial geomorphology is well-known, but biotic responses of insects remain poorly known. Investigation of experimentally-immersed wood in Australia shows native timber supports enhanced biomass compared to alien woods in open riverine areas or alien pine forests. Structural modifications of immersed wood by grooving and tunnelling of elmids encourage proliferation of surface-dwelling invertebrates. A diverse 'mining' fauna develops within immersed wood, including specialist caddis, beetles, and especially chironomid midge larvae. Recent discoveries of unusual midges in Australia and south-east Asia allow speculation about the evolution of immersed-wood mining. The subfamily Orthocladiinae contains many unrelated taxa with larval mouthpart modifications associated with mining. In contrast, in the subfamily Chironominae, wood-mining has not been associated with morphological differentiation and is restricted to fewer clades, but radiation is evident in several early-branching clades. Wood mining in each subfamily is inferred as ancestral and may be associated with differential survival from cataclysmic extinction events.

Grazing effects on arthropod distribution & abundance mediated by habitat structure & consequences for meadow pipits in the Scottish uplands

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Bird populations have significantly declined in upland landscapes of Britain in recent decades. The progressive increase in grazing herbivores has been implicated. The mechanism is uncertain, whether habitat structure indirectly affects birds via arthropod abundance and distribution or directly affects breeding birds in terms of nest site suitability or restricting the access of foraging birds to arthropod food. The GRUB (GRazing and Upland Birds) project is using an experiment located in the Southern Highlands of Scotland to investigate the effects of different grazing regimens on plant-insect-bird interactions. Vegetation, arthropods and Meadow pipit populations are being studied in a grazing experiment (4 grazing treatments replicated in 6 blocks) located in the Southern Highlands. Here we focus on the distribution and abundance of arthropods prior to the experiment and their short-term response to the varied grazing regimens of the experiment. We hypothesise that initial responses of arthropods in the wet, heterogeneous biotopes will be to structural changes in vegetation and that these will be related to the foraging preferences of sheep and cattle. We also explore how the distribution and abundance of arthropods relates to the position of nest sites of Meadow pipits.

The effects of stand age & silvicultural treatment on beetle biodiversity in coniferous stands in south west Nova Scotia

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Old-growth forest (OGF) in Nova Scotia typically has uneven-aged, multi-layered stand structure and contains much coarse woody debris. These traits provide microhabitats that support a large proportion of Nova Scotia's biodiversity. Many species, including invertebrates, depend on the deadwood substrates typical of OGF. Saproxylic beetles (family Coleoptera) depend on dead or decaying wood. They play a large role in nutrient cycling and decomposition in forest ecosystems. The goal of this study is to determine the relationships between forest stand age, silvicultural treatment, dead wood, and beetle diversity. Saproxylic specimens were gathered using four methods: pitfall traps, funnel traps, sweep-netting and manual grubbing. For the pitfall and funnel traps, effort and time were controlled; however, the sweep-netting and manual grubbing were only controlled for time. At present, we have completed a simple analysis of variance for both forest stand age and silvicultural treatment and have found that only silvicultural treatment has an effect on species richness. As well, we have identified several beetles previously unrecorded in Nova Scotia. The study is still underway; by August 2004, however, all results will be ready for presentation at the conference.

The systematics, biogeography & evolution of Tasmanian cave fauna

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The Australian island of Tasmania is richly endowed with caves in numerous isolated karst areas. These harbour diverse faunas including obligate subterranean terrestrial and aquatic species. Located at the southern latitudinal limits of the Australian continent, and mountainous over much of its area, Quaternary climate change has profoundly influenced geomorphic, hydrologic and biologic processes within the karst and caves. Many karst areas were directly affected by glacial ice and other cold climate processes, during multiple phases of Pleistocene glaciation. This paper presents a broad overview of the systematic composition and biogeography of the Tasmanian cave fauna, and critically examines the influence of Pleistocene climatic changes and glaciation in shaping the evolution of the fauna. The terrestrial obligate cave fauna is derived from lineages associated with cool and moist temperate forest habitats, including Coleoptera (Carabidae), Arachnida (Araneae, Opiliones, and Pseudoscorpionida), Diplopoda (Dalodesmidae) and Isopoda (Oniscidea). Cave-limited species within these groups have restricted distribution ranges that are often limited to a single karst area. Isolation of ancestral populations in caves, and their subsequent speciation, may have occurred during periods of cold glacial climate when forest cover retreated from karst areas.

Australian cave-dwelling planthoppers: relics or explorers?

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The Australian planthopper taxon *Solonaima* contains epigean as well as cave-dwelling species. Hence it is an ideally suited model system to study cave-adaptation. Assumably cave-inhabitation happened within *Solonaima* several times. This would support the adaptive-shift hypothesis after Howarth 1986, which regards cave-adaptation as an active process driven by the exploitation of a previously unused habitat and/or food resource. In case of the cixiid genus *Solonaima* this means feeding on roots in the adult stage, even though this feeding strategy is in epigean Cixiids only known for nymphs. The allopatrical distribution of *Solonaima* (epigean species occur in coastal rainforest habitats while cavernicolous species are restricted to Queenslands Outback) on the other hand supports another hypothesis. This relict hypothesis after Barr 1968 postulates that cave-adaptation is due to decreasing conditions on the surface, though that populations could have only survived in caves. Current data based on phylogenetic reconstructions make a synthesis of the two hypotheses plausible. The most likely evolutionary scenario is that some *Solonaima* species have undergone an adaptive-shift, though that they spent their whole life cycle in caves. During the desertification of Australia in the Miocene the surface-dwelling species in the dry areas became extinct. Only those who there switched to a permanent life underground survived.

A survey of biodiversity in Siahkal forests North Of Iran

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The forests of north of Iran are of the most important broad leaf trees of the word. The purpose of this study is to investigate tree diversity in the forests of Siahkal (north of Iran) with an area of about 13000 hectar. For this purpose, 18 sample plots 0.5 hectar have been randomly chosen and surveyed in different altitudes of Siahkal forests . The results indicated that with increas in altitude from sea level, species richness decrease while species diversity and evenness continue to rise. Using the statistical test of chi-square, a comparison of the observed and expected frequencies of different characteristics of the trees indicated that the difference was highly significant as far as dominant trees and the quality of their trunks in a cylenic position is concerned. As for the suppressed trees, different tree crowns, as well as curved trunks no significant difference was observed. From a regeneration point of view, the best case was observed at the middle altitudes (1300-1600 meter).Besides, because of existence of wandering animals at the altitude range of 1300 meter below sea level and 1600 meter above, the regeneration of trees are not considered to be desirable.

Emergence collection – a trial of biodiversity composition & longitudinal patterns of aquatic insects emergence in small rivers in Palawan, the Philippines

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This study presents results of the first emergence trap samples from streams in the Philippines and Greater Sunda. It compares aquatic insect emergence of two 4th order rivers and describes longitudinal patterns including estuaries. A decline of total emergence with increasing stream order was observed, affecting all major orders. Diptera, namely Chironomidae, dominated all sites. High abundances in Ceratopogonidae, Odonata, and Coleoptera were found, compared to other emergence studies from tropical and temperate latitudes. Ephemeroptera displayed a highly variable contribution to the emergence from Palawan either supported by the appropriate conditions for certain functional groups or limited by environmental variables such as pH. Trichoptera were likely to tolerate a wider range of environmental conditions and are consequently able to fill further niches where Ephemeroptera are under-represented. Except for the scarce abundance of Plecoptera no substantial differences in emergence composition at order level existed between temperate and tropical rivers, however, with a remarkable local variation. The results obtained with emergence traps which were modified for the tropics displayed high species richness for the most insect orders. This fact and general characteristics of emergence trap samples suggest a high potential of this method for qualitative-ecological, taxonomic, and biodiversity studies in the tropics.

East meets west: phylogeny & biogeography of the Polynesian thomisid spiders

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Spiders of the family Thomisidae are ambush predators that mimic the coloration of different substrates. Thomisid spiders occur on a number of remote archipelagos across the Pacific, having attained their highest diversity in the Hawaiian Islands. The twenty Hawaiian species are exceptionally diverse in coloration, reflecting mimicry of different substrate types. In contrast to the high species diversity exhibited by Hawaiian thomisids, a single species is known from each of three neighboring Polynesian archipelagos. Though relationships among different Hawaiian lineages have been extensively studied, the evolutionary history of biotas occurring on other Polynesian archipelagos and their relationship to the Hawaiian biota is poorly known. Here we reconstruct phylogenetic relationships among thomisid spiders occurring across Polynesia, including multiple islands from the Hawaiian, Marquesan, Society and Austral archipelagos, and outgroups based on mitochondrial and nuclear DNA sequences. Our results indicate that the Hawaiian thomisids are closely related to species in the genus *Misumenops* occurring in the nearest island archipelagos of the Marquesan and Society Islands as well as those that occur in North America. However, thomisids from the Austral Islands appear more closely related to those occurring in Western Polynesia, suggesting that thomisids colonized Polynesia from two opposing directions.

Comparative phylogeography of saproxylic invertebrates

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To conserve evolutionary processes within taxa as well as local co-evolutionary associations among taxa, habitat reservation needs to take account of natural genetic-geographic patterns. While vertebrates tend to have at least moderate dispersal and gene flow on a landscape scale, there are good reasons to expect many small invertebrates to be strongly subdivided. Saproxylic (rotting log) invertebrates are particularly likely to exhibit fine-scale population structures owing to their ecological specialisation and limited dispersal. It is a major challenge in conservation biology to develop efficient tools and model systems to understand genetic and evolutionary spatial patterns in less mobile, habitat-specific organisms. Further, it is necessary to investigate the relative importance of local adaptation and historical differentiation. Comparative phylogeography provides a framework for differentiating species-specific and habitat-specific processes, yielding insights into forces driving the geographic distribution of biodiversity in individual species and regional biota. I present an overview of collaborative research into an exciting model system centred at Tallaganda State Forest NSW. In this system, velvet-worms, springtails and terrestrial flatworms show unexpected levels of local genetic subdivision and highly congruent phylogeographic breaks. These animals offer a fine-scale benchmark against which co-distributed animals can be compared. I outline our current and future investigations.

The effects of forest type & dead wood composition on saproxylic beetle assemblages on logs placed in boreal forests

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Natural old-growth boreal forests contain large volumes of the coarse woody debris (CWD) and management practices have led to a reduction in both the volume and quality of CWD. Saproxylic beetles, many of which are red-listed, are dependent on such habitats. We tested the effect of forest management on assemblages of saproxylic beetles on logs placed in ten clear-cuts, managed forests and reserves in northern Sweden. Sites were surveyed to determine the volume and quality of CWD habitat available to saproxylic beetles in the surrounding area. Clear-cuts and managed forests clearly differed from reserves in most aspects of CWD composition and quality, in particular tending to have less wood in the later decay and larger size classes. While beetle assemblages in clear-cuts differed from those in reserves, assemblages in managed forests did not. In the first year of the experiment, assemblages in reserves and managed forests were dominated by the bark beetles *Trypodenon lineatum* and *Hylurgops palliatus* and other species dependent on the early stages of wood decay, and it is likely that any differences between the older forest types will become more apparent when the logs reach a later stage of decay.

Conserving Queenslands invertebrates

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The Queensland entomological community has been making a very important contribution to the conservation of invertebrates within the State. A range of programs has been developed to allow for formal cooperation between Queensland Environmental Protection Agency and entomologists, both essional and non-essional. BugCatch is one of our major programs. Volunteers from the entomological community assist us with surveys on National Parks and State Forests. Products from these surveys include species lists as well as important recommendations for fire and other management regimes that will assist in conserving the species within protected areas. Scientific purposes permits have been a long-term issue for entomological collecting due to their non-specific nature and difficulties with assessment by people without adequate knowledge of invertebrates. The permit assessment has been significantly improved with issue of Scientific Purposes Permits to representatives of the Entomological Society of Queensland. The permits are endorsed for members of the Society. The Queensland entomological community has assisted the Queensland EPA because we have been able to engage them in our conservation activities. This has occurred as a result of enthusiasm of a number of staff in he Department and the leadership provided by Don Sands.

Detecting the effects of environmental change above the species level using beetles in a fragmented upland tropical rainforest landscape

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Significant changes to invertebrate communities resulting from environmental change have been widely documented at the species level. However, measuring environmental changes at the species level requires significant taxonomic expertise and time. Using classifications above the species level such as family, or trophic group could fast track this process. However, questions remain whether such data possesses the required sensitivity. The Atherton Tableland landscape in northeastern Queensland, Australia, is composed of numerous rainforest fragments of varying size surrounded by cleared habitat (pasture), thus representing two scales of environmental change. The extreme, habitat loss, and the more subtle change associated with habitat fragmentation. We sampled almost 20,000 beetles from six replicate sites of pasture, small rainforest remnants, and both edge and interior from large rainforest remnants. We then compared beetle species, family, and trophic group patterns. Not surprisingly all datasets showed significant differences in community composition between pasture and all rainforest sites (ANOSIM $P<0.001$). Within rainforest sites, species data showed a complex pattern consistent with climatic variation of sites and a microclimate mediated edge effect, affecting small remnants. However, these same patterns were only weakly evident for the family and trophic group data. It is suggested that data above the species level may not have the sensitivity required to detect subtle environmental changes.

Arthropods in Tasmanian ***Eucalyptus obliqua*****: how unique is the canopy fauna?**

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Tasmanian native eucalypt forest canopies are host to numerous arthropod species, but most of what we know about this fauna relates to the minority of species, particularly leaf-feeders, liable to cause economic damage. Sustainable management of these forests requires understanding how associated biodiversity is partitioned in space and time. For instance, how important for arthropod biodiversity conservation is it to maintain old living trees in a forestry landscape dominated by younger trees; and how distinct is the fauna of living trees when compared with that of coarse woody debris? A series of studies at the Warra long-term ecological research site in southern Tasmania is beginning to address this knowledge gap for the commercial species Eucalyptus obliqua. This paper discusses these studies, and presents some preliminary findings about the degree of overlap in arthropod faunas among different strata and microhabitats from the canopy to the forest floor, with particular emphasis on saproxylic species.

A research agenda for insects & dead wood

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In recent years, great advances have been made in our knowledge of the ecology, evolutionary biology, biogeography and management of saproxylic insects and their dead wood habitat. There is now a greater level of awareness of the need to cater for their requirements in today's managed forest landscapes. However, having raised the profile of saproxylic insects, we are now being called upon to translate our knowledge gains into management principles. This throws up new research challenges, many of which can only be addressed through adaptive management. This paper outlines and prioritises some of those challenges.

Scale insect radiations on eucalypts

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At least 200 species from more than 50 genera belonging to a total of 10 families of scale insects (Hemiptera: Coccoidea) feed on species of *Eucalyptus sensu lato* (including *Corymbia*) in Australia. Most (about 60%) of these species belong to the Eriococcidae and approximately 80% of these eucalypt-feeding eriococcid species induce complex galls on their host plants. Molecular phylogenetic analysis of the Eriococcidae reveals two unrelated, speciose clades of gall-inducing taxa associated with eucalypts. One clade comprises the genera *Ascelis*, *Cystococcus*, *Lachnodius*, *Opisthoscelis* and relatives, and the second is the extraordinary genus *Apiomorpha*. Gall-inducing eriococcids are more host-specific than their non-galling relatives and, at least in *Apiomorpha*, appear to be cospeciating with their hosts.

Maculinea Butterflies of the Habitats Directive & European Red List as Indicators & Tools for Habitat Conservation and Management

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Insects have declined far more rapidly in modern European landscapes than any other known taxa. As agricultural intensification and abandoned hay meadows threaten European grassland biodiversity, we are assessing the suitability of *Maculinea* butterflies as bioindicators. The EU-funded project MACMAN (EVK2-CT-2001-00126) aims to develop standards for monitoring the 5 species of *Maculinea* butterflies as indicators and tools for grasslands and their management. With partners from 19 countries (including Associated partners) research is being conducted throughout the entire European range of each species, encompassing different biogeographical conditions and population structures. Trophic and functional relations and genetic biodiversity of the *Maculinea* are being investigated, population structures (from isolated to metapopulations) are being analysed, MVP and MVM thresholds established, co-occurrence data analysed and habitat models derived. The scientific questions answered during the project should provide guidelines on the management of grasslands for high biodiversity for land users and managers. We will provide users in EIA (Environmental Impact Assessment) with simple robust tools for assessing the effect of land use changes on the persistence of endangered communities of wildlife, which will involve a unique standardisation of approaches and applications across Europe. Subcontractors (i.e. National Parks and EIA consultants) will test our recommendations.

The role of dead wood in maintaining forest diversity

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Dead wood is one of the major components of forests that contribute to overall diversity primarily through insects that feed directly on or in it. Likewise, a variety of organisms benefit by feeding on those insects. What is not well known is how or if dead wood influences the composition of the arthropod community that is not solely dependent on it as a food resource and whether woody debris influences prey available to generalist predators. One group likely to be affected by dead wood is ground-dwelling arthropods. We have studied the effect of adding large dead wood to unburned and frequently burned pine stands to determine if dead wood was used more when the litter and understory plant community are removed. We have also studied the effect of annual removal of dead wood from large (10 ha) plots over a 5 year period on ground-dwelling arthropods. In related studies, we have examined the interrelationship of an endangered woodpecker that forages on live trees, its prey, and dead wood in the forest. The results of these and other studies show that dead wood can influence the abundance and diversity of the ground-dwelling arthropod community, and prey available to generalist predators not foraging directly on dead trees.

Effects of two genetically controlled chemotypes on the community structure of herbivorous insects

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Salix sachalinensis Fr. Schm (Salicaceae) has two distinct chemotypes: one biosynthesizes ampelopsin as a major component of low molecular weight phenolics in their leaves (A-type), and the other biosynthesises -D-glucopyranose 1-*trans*-*p*-coumarate and -D-glucopyranose 1-*trans*-cinnamate in addition to ampelopsin (AP-type). These two chemotype are genetically controlled. We investigated the effects of these chemotypes on the community structure of herbivorous insects. We placed potted clones of two chemotypes at a field study site located in the flood plain of Ishikari river in Hokkaido, northern Japan, and examined the plant leaf traits and herbivore communities of the cloned chemotypes. There were significant differences in leaf phenolics between the two chemotypes. Total abundance, species richness, and species composition of herbivorous insects did not differ significantly between the chemotypes, whereas the relative abundance of herbivores differed significantly. In particular, the density of the willow leaf beetle, *Plagiodera versicolora*, was significantly higher on the A-type than on the AP-type, In contrast, the density of leaf galls was significantly lower on the A-type than on the AP-type. This distribution pattern may indicate a potential ecological cost of leaf phenolics production in *Salix sachalinensis*.

The fine-scale physical attributes of coarse woody debris associated with utilization by ants (Hymenoptera: Formicidae)

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Coarse woody debris (CWD) is increasingly recognized in Canada for its contribution toward biodiversity. Wood, which has low specific heat, provides a thermally favourable environment in this cool climate. Ants contribute to physical breakdown of wood, and colonies are a significant food source for vertebrates like grizzly bears and woodpeckers. Where CWD retention has been a management objective, volume is usually used as the sole indicator. However, CWD differs greatly in quality between non-harvested and harvested sites. Pieces remaining after harvest are shorter, thinner, lacking in bark, and missing decay classes typical of non-harvested sites. This has led to the concern that post-harvest CWD may not be biologically functional. In this study, conducted in cool moist sub-boreal forests, we are examining the fine-scale physical attributes of CWD utilized by ants, in non-harvested and harvested sites (8-10 years post-harvest). Results to date indicate significant effects of stand structure and climate on ant ulitization of CWD, with only a few species tolerating cool interior stand conditions. The possible significance of regional climate, insolation, temperature, and moisture will be discussed.

The importance of type (logs, snags or the top of trees) & treatment (shading or burning) of dead wood for its attractiveness to saproxylic beetles

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Intensive forest management resulting in reduced fire frequency, the amatic decrease of dead wood and changes in tree species composition in boreal forests is probably the main reason for the amatic decrease of many saproxylic species in Fennoscandian boreal forest. This has lead to changes in forestry practise, including dead wood retention, which value not yet has been rigorously evaluated. To evaluate the value (from a conservation perspective) of different types of dead wood (logs, snag or remaining top of the tree) and find ways of improvement (burning and shading of logs) we performed a large-scale field experiment in northern Sweden. Species richness and the abundance of saproxylic coleopterans were higher on logs than on snags and tops but the different types of dead wood also attracted different beetle communities. Some red-listed species as the endangered *Monochamus urussovi*, was most common on tops and also found on snags, but not on untreated logs. Treatment of logs also influenced their suitability for red-listed species. Eleven, seven, five and four red-listed species was found on burned, control, shade net and naturally shaded logs, respectively. The implication of these findings for conservation strategies is discussed.

Effects of urbanization on insect-plant interactions on dominant & invasive plant species in Sydney, Australia: testing enemy release & plant stress hypotheses

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Declines in dominant native vegetation in urban remnants are often associated with the establishment of invasive species beyond their natural range. The "enemy-release" and "plant stress" hypotheses are often offered to explain these patterns; the former reflecting the loss or absence of natural enemies in urban environments and the latter reflecting high levels of nutrient enrichment caused by urbanization. We examined the interactions between soil nutrients, plants and the arthropods associated with them in urban forest remnants in Sydney, Australia. *Angophora costata*, the dominant tree in these remnants, supported a different assemblage of arthropods and suffered higher levels of herbivory in urban areas where soil nutrient levels were high. Manipulative experiments also showed that *A.costata* seedlings could not establish in high nutrient soils, supporting the contention that enemy-release and plant stress contributed to its decline in urban remnants. A major native weed *Pittosporum undulatum* showed contrary patterns, suffering less herbivory in urban areas and no response to nutrient enrichment, suggesting that its success in new environments may be partially attributed to release from natural enemies. By driving biodiversity loss and the modification of soil properties, urbanization disrupts fundamental ecological processes and the ecological integrity of urban remnants.

Ecological comparisons within the arid zone

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The subterranean life on the eastern seaboard of Australia is often closely coupled with surface relatives and influenced by quite recent events, such as Pleistocene glaciation. In contrast, the subterranean fauna of the Australian arid zone is characterized by relictual faunas in which surface relatives are now far distant in space and time. Still living in subterranean ecosystems, they comprise samples of the fauna 'snatched' from those of various geological Periods. Within the subterranean realm of Australia are clear signs of past connections with the various supercontinents and the Tethys ocean, and clear signals from major marine transgressions, as well as from the spread of aridity during the Tertiary. As such the subterranean realm has preserved ancient biodiversity of extraordinary richness and still largely in a pristine state. Three distinct faunas are discussed a) the terrestrial cave fauna of northwestern Australia dominated by relictual rainforest faunas having both temperate and tropical elements; b) the aquatic fauna of the northwestern near-coastal (anchialine) groundwater dominated by a fauna with Tethyan affinities whose closest relatives inhabit similar ecosystems on either side of the North Atlantic; c) an aquatic fauna inhabiting isolated 'island' aquifers in the desert.

Evolutionary diversification of an ant-plant-scale insect tripartite symbiosis in Southeast Asia

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Many tropical plants house ants and thereby gain protection from herbivores. Most of these myrmecophyte-ant systems include scale insects as the third associate. Scale insects live inside the plant hollow stem, suck the plant sap, and provide honeydew to the ants. We investigate the evolution of the three-way interaction among myrmecophytic *Macaranga* plants, their inhabitant *Crematogaster* (*Decacrema*) ants, and the trophobiont *Coccus* scale insects. Previous studies show that these three genera have a tight association although a few other genera are sometimes involved. The mtDNA COI phylogenies of comprehensively-sampled ants and scales reveal ten and five well-supported lineages, respectively. Comparisons of the two phylogenies and a published *Macaranga* phylogeny indicate an absence of cospeciation. Whereas the ant-scale and plant-scale interactions are diffuse, the ant-plant one is specific in the sense that host shifts have been constrained by plant stem traits that are themselves correlated with *Macaranga* phylogeny. Applying COI divergence rates of 1.5% per My, the *Decacrema* ants and the *Coccus* scales from *Macaranga* are inferred to be at least 12, and 10 My old, respectively, indicating that they have been diversifying contemporaneously since the mid-Miocene in the wet forest community of Southeast Asia.

Endemicity rates in the West Indian hotspot

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The islands of the West Indies have recently been recognized as a Hotspot of global biodiversity, but the size and distribution characteristics of the insect fauna remain poorly documented. The view of regional vs. single-island endemism is greatly effected by lack of collecting, misidentification, and inadequate revisionary studies. Using beetles as an exemplar taxon, island faunal inventories as well as single-taxon revisions show the endemicity rates on small islands to be far higher than currently expected. Although rates are highest between island groups separated during the Pleistocene, even islands that have had an independent identity only since the eustastic rise at the end of the Pleistocene harbor endemic elements. Discovery rates on islands in the Northeastern Caribbean document the lack of comprehensive knowledge of the regional fauna and the underestimation of both total species and endemicity that result from this ignorance. Concentration on the faunae of large islands may significantly under-estimate total biodiversity in insular faunae.

Ethological interaction between whiteflies (Aleyrodidae : Hemiptera) & ants in certain locales along the Eastern & Western Ghats of India

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The Eastern and Western ghats of India are considered to be rich in biodiversity in India and the latter is recognized as one of the eighteen biodiverse hot spots of the world. During a survey for whiteflies in these regions a number of species of this group of insects have been collected and identified. Exudation of honey dew by whitefly nymphs attracts several ant species. Of the many species of whiteflies recorded along the ghats of India, the genera *Rhachisphora, Tetraleurodes* and *Aleuromarginatus* have been found to be having mutualistic interaction with 16 ant species. The number of ants tending whitefly on a leaf is positively related to the number of whitefly on that leaf. The activity pattern of each species of ants had its own time period and the activity time varied at different time intervals. Ants took more time for antennal grooming on the whitefly compared to palpating the honey dew with its mandibles. Large sized ants attended to more number of whitefly nymphs and spent longer time for feeding while smaller ones shared the exudation with its own members and also with other species. The ant attended whiteflies faced minimum parasite attack compared to non-ant attended whiteflies.

Beetle attraction to volatiles from fruit bodies & mycelium of wood decaying fungi

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The interactions between beetles and wood-fungi are important for conservation of saproxylic beetles. Beetles living in fruit bodies of fungi are known to be attracted to volatiles from the fungi but it is not known if some beetles also are attracted to smell from the mycelium. The aim of this experiment was to test the attraction effects of volatiles from fruit bodies, mycelium and burned wood on beetles. In a randomized block design, nine substrate types - burned, *Fomitopsis pinicola* fruit body, *F. pinicola* mycelia, *Fomitopsis rosea* fruit body, *F. rosea* mycelia, Control, *Phellinus chrysoloma* fruit body, *Phlebia centrifuga* mycelia and wood - were attached separately to window traps in four old growth spruce forests in northern Sweden. Substrate type influenced both abundance and species richness of beetles and mycelium and fruit bodies attracted different species assemblages. Highest abundance of beetles occurred on mycelia of the red-listed *F. rosea* and fruit bodies of *F. pinicola*. The bark beetle *yocoetes autographus* preferred mycelia of *F. rosea* and the rove beetle *Lordithon lunulatus* showed preference for fruit bodies of *F. pinicola*. The results indicate that conservation of rare fungi species is important for many beetle species and beetles seem to be able to find suitable substrate using volatiles emitted from the mycelium.

Conservation of beetles, coarse woody debris & Natura 2000 in Europe – Slovenian experience

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The author presents the inclusion of elements of nature protection based on the European network Natura 2000 in the university education programme of forestry and in forestry practice. With the entrance into EU Slovenia is accepting, in the field of nature protection, the relevant legislation of the Community. Nature protection is based on Council Directive 92/43/EEC (the Habitat Directive) and Council Directive 79/409/EGS. In Slovenia the definition of potential Sites of Community Interest for the species of Coleoptera listed in Annex II of the Habitat Directive is nearing the end. 13 species from Annex II and another three from the list proposed by the candidate member states are present in Slovenia. 11 of the listed 16 species are saproxylic, two are water living species, one is a hygrophylic species, one is a cave dweller and one is a soil dwelling species. All except one live in forest habitats. They are mostly stenotopic species, dependent on old trees, above all on conifers or deciduous coarse woody debris. The characteristics of particular species, the methods of their protection and the role of particular species in the forest ecosystem are presented.

Endemism of dragonflies & damselflies (Order: Odonata) in Western Ghats of Kerala, India, & the constraints in their conservation

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India contributes about 500 species of dragonflies and damselflies to the total of 5600 species of world population. And 115 species are endemic to this country. Fraser (1924,1933-1936) who had extensively studied the Odonata fauna of this region, made a base line data of dragonflies and Damselflies of this area. A good number of Odonates are endemic to Western Ghats and neighbouring states (67 out of 177). Western Ghats is an array of mountains with evergreen tropical rain forests. Moreover, as far as biodiversity is concerned, Myers *et al* (2000) described Western Ghats as one of the "Hottest hot spots" of the world. But, high human population density (819 people/sq.km) in Kerala state leads to various anthropogenic activities such as migration of people to mountain/forest side for housing, construction of hydroelectric projects, change in land use for plantations such as Tea, Coffee, Rubber, Cardamom etc. Besides, the usage of very high quantity of insecticides and fertilizers in these plantations not only pollute the aquatic ecosystems in the mountain regions, which seriously affect the breeding of rare odonates, but also destroy the lower water bodies through riverine systems which carry high rate of these organic matter, and results in threatening the very existence of the rare endemic species of dragonflies and Damselflies of this region.

Ants as indicators in the quest for landscape thresholds in southern Queensland poplar box woodlands

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Broad scale clearing of remnant vegetation for agricultural development can lead to habitat loss, fragmentation and disruption of important ecosystem processes. To investigate these issues, this project, funded by LWA Australia and Queensland Department of Natural Resources and Mines, aims to define and quantify ecological thresholds for habitat retention and management of poplar box woodlands in southern Queensland. A suite of environmental markers of ecological condition, genetic diversity and vegetation and soil health is being utilised to determine indicators of status and condition, measures of viability and, where possible, threshold values. The study area extends across 4.5 million hectares in the Queensland Murray Darling Basin. Data were collected from sixty *Eucalyptus populnea* remnants and regrowth patches, of which twenty were studied intensively. Data were analysed against landscape metrics reflecting different management regimes. Land use of the surrounding matrix, distance to neighbouring remnants and historical disturbance (1988 to present) were also determined. Ants are recognised as significant indicators of habitat condition. Pitfall trapping for ants took place three times at the intensive sites. Differences in ant assemblages, in terms of diversity and functional groups composition, were compared across sites and against changes in landscape pattern using multivariate analyses.

The arthropod fauna of cool temperate *Nothofagus*-dominated rainforests in Australasia: the first site

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A transect of one-hectare reference plots is proposed within the chain of remaining patches of microphyll and notophyll fern forests dominated by *Nothofagus moorei* and *N. cunninghamii* in Australia. These forests are relictual and considerably simpler than the contiguous warm temperate and subtropical rainforests of the east cost of Australia. The transect will be extended beyond Australia as the opportunity presents itself. The first, developmental, plot has been established in Lamington National Park, south-east Queensland. Following a total botanical inventory and establishment of permanent botanical subplots, wet season arthropod surveys using pitfall, interception, and yellow pan traps, plus litter, tree bark and canopy knockdown samples have been carried out. Early results will be presented. Even at the ordinal level the samples show amatic differences from otherwise comparable samples from subtropical forests at the same latitude. Explanations for these differences include physical differences in forest structure, altitude-related climatic contrasts, and the biogeographic history of insect/plant associations in these relictual ecosystems. Family and morpho-species results for the Coleoptera and Heteroptera are also presented. Exactly comparable surveys will be carried out at three more southerly, NSW locations, plus sites in Victoria and Tasmania. Comparisons with existing results from New Zealand and Chilean forests will be made.

The effects of fire severity & post-fire logging on boreal ground beetles (Coleoptera, Carabidae)

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Fire is one of the most important natural disturbance forces in boreal forested ecosystems. Yet, post-fire logging and fire-severity effects on beetles are poorly studied. We collected ground beetles by pitfall traps in 2003 in central Alberta, Canada, from 384 mixed-wood (Aspen, White Spruce) sites at House River, a ca. 3000-km2 area burned in May 2002. The sampling sites varied in burn severity and logging intensity. We present preliminary results of our study, with a general aim to determine the effects of post-fire logging and fire severity on ground-beetle assemblages.

Parataxonomy versus taxonomy in biodiversity studies – pitfalls & applicability of 'morphospecies' sorting

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In biodiversity studies, sorting of samples to parataxonomic units (RTUs, morphospecies) is generally considered a sufficiently reliable and conservative approach. It is obviously time-saving because it avoids the burdens of taxonomy. However, evaluations of parataxonomic sorting by taxonomic resorting show many overestimations of species numbers and is, therefore, not necessarily conservative. Sorting errors can be more than 100% (median in the present compilation: 22%). Even if the cumulative results for diverse groups like beetles have a low overall error, the error rate within the families is generally much higher, causing misleading results in multivariate analyses. Since the error rate does not depend only on the group to be sorted, but also on the sorter and the sample, the sorting error is not predictable. However, if results of parataxonomic sorting show clear and biologically meaningful patterns, the sorting is likely to be reliable. Weak or no detectable patterns may easily be caused by erroneous sorting. I'll discuss in which type of studies parataxonomic sorting results can be used and in which they are useless.

Evaluation of efficiency & taxonomic bias of the Winkler method for soil macro-invertebrate extraction

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The Winkler method is a cheap and simple method to extract macro-invertebrates from litter and soil, but has never been comprehensively evaluated. We ran the extraction for seven weeks, collected the extracted invertebrates 18 times and completed the experiment by handsorting of the soil/litter (from an English woodland). We identified the minimum extraction times for invertebrate groups, ranging from 2 days for ants, 3 days for adult beetles up to 3 weeks for earthworms and 4 weeks for centipedes. The method is not efficient for woodlice and molluscs. We propose coefficients to estimate the real number of specimens for each invertebrate group from samples obtained after suboptimal extraction periods. To identify taxonomic bias related to extraction time, we checked beetles and centipedes at family and species level. The rank-abundance order of the six most abundant beetle families remains unchanged from three days onwards. The final rank-abundance order of centipede species, however, was reached only after 15 days. Traditional short extraction times of 2 or 3 days might be sufficient for ants and beetles, but not for getting an accurate picture on the relative abundance of most other soil macro-invertebrates.

Evolutionary radiation of stiletto flies in Australia

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The cosmopolitan lower brachyceran family Therevidae, stiletto flies, have nectar feeding adults and soil-dwelling predatory larvae. Intensive field and systematic research over the last decade now estimates the Australian fauna at 700 species. Phylogenetic studies, including molecular and supertree analyses, show three independent lineages, the *Taenogera* group of genera (150+ spp.), the *Anabarrhynchus* group (200 spp), and the *Agapophytus* group (250+ spp). Large numbers of superficially similar species occur in each lineage and each has a small number of species in Chile or Argentina. Australian therevid alpha diversity is remarkably high, with over 120 species known from sites such as the Warrumbungles where over 8700 specimens have been collected. Field research suggests that faunal turnover (beta diversity) is almost complete over a few huned kilometres in both eastern and western Australia. Why are there so many stiletto flies in Australia? We believe that speciation in these therevid evolutionary radiations has occurred through the Tertiary in response to the increasing aridification of Australian environments, causing changes in the microhabitat requirements of larvae and adults. This gradual aridification has occurred simultaneously with alternating xeric and mesic periods, causing expansion and contractions of species distributions and speciation by allopatry.

Spiders, spinifex, rainfall & fire: factors influencing spider diversity in an Australian desert

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Critical to conservation of biodiversity in Australia's arid zone is an understanding of the spatial and temporal patterning of biota. For mammals and reptiles, both rainfall and fire have been found to be forces determining these patterns. Few studies however have considered the influence of these factors on invertebrates. Fewer still are studies where there is a long-term data set available to consider responses over an extended time frame. This talk describes the results from spiders collected using pitfall traps over a period of 14 years (1989- 2003). It was completed as part of an experimental fire study conducted in the Great Victoria Desert, Western Australia. Rainfall was found to be the dominant driving force in this system with fire having a lesser effect. These finding's reinforce the importance of long-term studies when examining complex ecological processes.

Maintaining saproxylic insects in extensively managed boreal forests

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Recent work on saproxylic insect assemblages in western Canadian boreal forests has demonstrated that diversity and variability are high, and that there are sampling and taxonomic challenges to assessment of these insects. Some major determinants of assemblage structure include tree species, degree of decay, stand age and cause of tree death. Experimental approaches have revealed the differential impacts of the stand-replacing disturbances of wildfire and harvesting on saproxylic insect assemblages. Exploration of saproxylic insect responses to variable retention harvesting and experimental burns is contributing to identification of optimal management prescriptions for boreal forests. Understanding of processes determining saproxylic insect diversity patterns and responses would benefit from increased attention to natural history. Such work would lead to a biologically meaningful classification system for dead wood to help to better identify habitats (and associated species) at risk due to forest management. This tool could also be used to improve strategies to better maintain saproxylic organisms in managed boreal forests.

Conservation of biodiversity in managed boreal forests

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In response to the increasing awareness of the importance of preserving biodiversity, forest management practices have been radically revised over the past few years. A crucial question is whether or not the "New Forestry" will actually achieve the goal of maintaining biodiversity. A major threat to biodiversity in Swedish forests is the decline of Coarse Woody Debris (CWD), which is an essential resource for many organisms and plays an essential role for the structure and function of boreal forests. Removal of CWD in commercial forestry has depleted important resources for many rare wood-living (saproxylic) beetles. Replenishment of CWD has been a prominent goal of the "New Forestry", through practices such as retention of living trees on clearcuts, creation of standing dead wood such as snags, and abstinence from removal of wind-felled trees. In an ongoing project in central Sweden we are studying the dynamics of CWD as well as assessing beetle species richness and composition in a landscape where the "New Forestry" has been applied. Results from the project will be presented.

Solitary bee communities in a vineyard landscape

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Because bees do not pollinate grapes, the conversion of oak woodland to vineyard may present an especially difficult problem for solitary bees. These bees may lose both nesting habitat and food resources when oak woodlands are converted to vineyards. The remaining wild habitat fragments in heavily converted landscapes like the Napa Valley of California, are the equivalent of island refuges in an inhospitable sea. To study the effects of these landscape-level changes on the solitary bee communities of the remaining oak woodlands, we developed criteria for matching sampling areas in vineyard-dominated landscapes with similar areas of oak habitat not dominated by vineyards, intensive agriculture, or development to create a randomized block design of 'treatments' (areas with high degree of 'vineyardization') and 'controls' (oak-dominated areas). We compare solitary bee species richness and abundance in these areas and floral host use. We find a significant difference in species richness and floral host use between treatments. In particular, non-native plant species appear to be a significant food resource late in the flowering season.

Speciation in natural test-tubes

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Theoretical models for sympatric speciation have become increasingly sophisticated, but empirical evidence remains restricted to a small number of vertebrate taxa. The plethora of subterranean diving beetles (Coleoptera: Dytiscidae) species that recently have been discovered in numerous isolated calcrete (limestone) aquifers in the arid inlands of Australia, might well be the first invertebrate taxa that can be added to this list. Currently, 70+ species have been recognized, each of them restricted to a single calcrete aquifer. Phylogenetic evidence, based on mtDNA, shows that besides numerous cases of allopatric speciation, resulting from isolation of several ancestral species in separate widely distributed calcrete aquifers, approximately 33 % of these aquifers contain pairs of species that are closely related. These sister species pairs also differ considerably in size and morphology. We investigate the mode of speciation for these sympatric sister species pairs by applying a simple model that simulates random occupation of niches by ancestral surface species during colonisation of the subterranean environments. We show that the number of observed sister pairs is better explained by a drive to occupy empty niches by species within the aquifer than by repeated colonisation of the aquifers by the same ancestral surface species.

Islands in the desert: evolution of subterranean diving beetles (Coleoptera: Dytscidae: Bidessini, Hydroporini) in arid Australia

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Isolated calcrete aquifers in arid inland Australia contain a rich invertebrate subterranean water fauna (stygofauna), comprising Amphipoda, Syncarida, Ostracoda, Copepoda and also the world's most diverse assemblage of subterranean diving beetles. Here we test whether the Adaptive Shift Hypothesis (ASH) or the Climatic Relict Hypothesis (CRH) is the most likely mode of evolution for the Australian subterranean diving beetles, by using a mtDNA phylogeny, linearized using a relaxed molecular clock method. We found that, 1) individual calcrete aquifers contain several diving beetle species of distantly related lineages and/or a single pair of sympatric sister species that significantly differ in size and morphology, 2) evolutionary transitions from surface to subterranean life took place in a relatively small time frame, and 3) most of the variation in divergence times of the sympatric sister species is explained by the variation in latitude of the localities, which correlates with the onset of aridity from the North to the South and with an aridity maximum in the Early Pliocene (5 Mya). Several lines of evidence inferred from the molecular clock analyses support the CRH, indicating that the evolutionary transitions took place during the Late Miocene and Early Pliocene as a result of aridification.

Wood, elytra, and antiquity: geologic & phylogenetic history of dead wood Coleoptera

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Ecologists use beetles as indicators because they are very species-rich, and are easy to collect manage in the laboratory. They are one of the most important groups of dead wood invertebrates, and may have been associated with rotting wood since their origin in the Permian (265 My). While studies in coarse woody debris (CWD) ecosystems are increasing, systematic studies of beetles are lagging: taxonomic revisions are needed for identification while phylogenies are needed to place groups of interest in an evolutionary context. The New Zealand CWD beetle fauna consists of approximately 5,000 species and up to 80% of the species are not described and/or need revision. Phylogenetic study of several Polyphagan groups shows that most CWD beetles are not necessarily basal taxa, and most belong to Gondwanan lineages. New Zealand taxa lineages have relatively long phylogenetic branch lengths suggesting that they are relatively old in comparison to their sister taxa located elsewhere in the world. Among mycophagous beetles, most host associations are primitive and may have predated the split between New Zealand and Gondwana. The CWD fauna could be one of the oldest in the world.

Rearing of *Sarcophaga penicillata* (Diptera: Sarcophagidae), for release as a biological control agent for *Cochlicella acuta* (Mollusca: Hygromiidae) in South Australia

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The conical snail *Cochlicella acuta* is a major grain contaminant in South Australia and is concentrated mainly on the lower end of Yorke Peninsula because of the region's limey soil type. Following an introduction from France and a quarantine testing phase over 2-3 years, the parasitic fly *Sarcophaga penicillata* was first released on South Australia's Yorke Peninsula in April 2000 for control of the conical snail *C.acuta*. *S. penicillata* has since established at several release sites. Laboratory rearing for release of *S.penicillata* was conducted at the Waite institute, South Australia. Two methods of rearing were tested and the *S.penicillata* parasitism rate in the laboratory colony ranged from 10-20%. In addition, experiments were conducted to investigate the pollen and nectar feeding habits of adult *S.penicillata* on Queen Anne's Lace (*Ammi majus*, Family: Umbelliferae) and Canola (*Brassica Napus*). This paper presents the recent developments with the laboratory rearing program and results from the 2003/04 fly release season.

Discriminant analysis of the disruptive impacts of a horticultural mineral oil & two synthetic pesticides on citrus ecosystems

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The impacts of an *n*C24 horticultural mineral oil (HMO) and two synthetic insecticides (carbaryl and methidathion) on the abundance and species diversity of soil-dwelling arthropods were evaluated in two citrus orchards on the central coast of New South Wales, Australia. In the first orchard, low (2,000 L/ha) and high (10,000 L/ha) volume sprays of HMO, carbaryl and methidathion were applied in treatments in summer 1996/1997 to mature Valencia orange trees (*Citrus ´ aurantium*). Arthropods present in leaf litter and surface soil samples taken from under trees were extracted with Berlese funnels. Results indicated that while the HMO had no significant impact on the arthropods, the synthetic insecticides, irrespective of spray frequency or spray volume significantly reduced the abundance and species diversity of the arthropods including springtails and the generalist predators, such as spiders, staphylinid beetles and gamasid mites. In the second orchard, HMO and methidathion sprays were applied to blocks of trees twice using an oscillating boom sprayer in February 1998. Results were similar to those observed in the first orchard. No significant disruption of soil-dwelling arthropods caused by HMO was observed. Methidathion was found to be significantly disruptive to most soil arthropods, but not to carabids or ants.

Reconstructing historical species associations among archival natural history specimens to evaluate endangered species status

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Specimens from the *Fauna Hawaiiensis* were associated by cluster analysis, thereby reconstructing historical assemblages of Hawaiian carabid beetles (Coleoptera: Carabidae) observed during the 19th Century. Specimen associations permit concise hypotheses of habitat preferences for many of the 32 carabid species collected during the early period of European scientific exploration (1874-1902) but not observed since. These species associations are diagnosable by ecological situations observable today. Suites of recently rediscovered species collected in sympatry during intensive faunal surveys starting in 1991 are clustered on the phenograms of historical associations, indicating concordance of ecological relationships between historical and present-day faunas. Absence of entire species clusters from recent collections suggests actions of common agents leading to extinction or extreme population reduction. Future consideration of official conservation status for Hawaiian carabid beetle species must account for the status of ecologically associated species, and the limited likelihood that individuals of extant species can be consistently observed in nature due to natural relative rarity or secretive habits within restricted geographic and ecological distributions. Historical specimen associations serve as the best guides for continuing efforts to monitor known faunal members and to rediscover long-missing species. These associations also serve to link information concerning individual species with particular habitats.

Vanuatu platynine Carabidae (Coleoptera) as the impetus for following cladistic tracks across the southwest Pacific

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The carabid beetle tribe Platynini includes numerous diverse lineages across a fragmented Laurasia, with associated radiations in South America, Africa and Southeast Asia. The tribes' limited diversification in Australia and the Pacific Islands is constrained by this Laurasian history, though speciose faunas inhabit New Guinea (125 spp.) and Hawaii (131 spp.). This contribution focuses on taxonomic characterization and relationships of the Vanuatu platynine fauna; a modest monophyletic radiation of five species, plus single representatives of five other clades. In order to define generic membership for these taxa, cladistic analysis including numerous taxa from throughout the southwest Pacific and Hawaii was carried out. The cladogram was robustly rooted using a salient character of the male reproductive tract, defining *Lorostema* Motschulsky—6 species ranging from Sri Lanka to the Philippines and Tahiti—as outgroup to the Pacific lineages. Cladistic area relationships define tracks that connect New Zealand and Fiji, and New Guinea and Vanuatu, with these two areas sister to Fiji. Hawaii is placed basal to the extensive radiation of Pacific clades inhabiting Melanesia and Polynesia. This early dating for the Hawaiian diversification is consistent with colonization of the Hawaiian archipelago well before the emergence of the oldest currently occupied island, Kauai.

The extended phenotype of aspen: consequences of variation for ecological interactions

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Trembling aspen (*Populus tremuloides*) is a dominant species of early-successional forests over much of North America. It is a primary food plant for over 100 species of herbivores, and can experience massive defoliation over extensive areas during outbreaks by insects such as forest tent caterpillars. It exhibits striking phenotypic variation in secondary chemistry, as a consequence of genetic, environmental, and G x E factors. Genetic variation in chemistry is related to a trade-off between growth and defense, and expresses itself as differential resistance to herbivores (invertebrate and vertebrate) as well as to pathogens. Aspen also exhibits genetic variation in tolerance to defoliation. Aspen chemistry can influence herbivore (e.g., forest tent caterpillar, gypsy moth) performance not only directly (bottom-up effects) but also indirectly, via impacts on insect natural enemies (top-down effects). Moreover, during outbreaks, herbivore-induced changes in foliar chemistry interact with genotypic differences in aspen to alter the quality of both green leaves for late-season folivores and leaf litter for detritus feeders. Differential resistance to, and tolerance of, defoliation, may contribute to a geographic mosaic of community assemblages and ecosystem function in landscapes dominated by aspen clones.

Trunk invertebrate faunas of Western Australian forests & woodlands: relationships with avifauna usage

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Trunk–associated invertebrates were sampled on marri, *Eucalyptus* (*Corymbia*) *calophylla*) along a transect from Karragullen, in the hills near Perth, through to yana, 150 km to the southeast. This represents a op in annual rainfall from 1078 mm down to 504 mm. Invertebrates were sampled by intercept traps, which collect animals that attempt to land on the trunks, and bark traps, which collect invertebrates that move, or live, on the trunk. The range and abundance of invertebrates sampled was generally greater in the intercept than the bark traps. Invertebrate abundance, activity and biomass on bark were strongly seasonal, with greater numbers being found during the moister periods. Arthropod levels tended to be greater on marri at the ier, eastern end of the transect, particularly on the three locations within the wandoo woodland. These trends are discussed in terms of soil nutrients, the rainfall gradient and plant community composition. Bark arthropods are an important food source for forest and woodland birds, with 15 to 25% of prey taken by insectivorous birds coming from bark substrates. As many as five species of birds in the southwest forests sampled are bark specialists, taking more than 40% of their prey from bark substrates. The frequency of bark-foraging and the number of bark-foraging specialists increased along the transect as the abundance and richness of bark arthropods increased.

Fire-killed trees provide habitat for arthropods & resources for vertebrates

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Dead trees are an unappreciated part of the environment and, in urban parks, are considered a safety hazard that must be removed. In 1989 and 1996, vandal-lit fires in Kings Park, Perth, killed large numbers of jarrah (*Eucalyptus marginata),* tuart (*Eucalyptus gomphocephala*, and sheoak *(Allocasuarina fraseriana)* trees. Trunk-associated arthropods were collected from live and dead trees of these three species in summer 1999 and winter 2000, using intercept and bark traps. Higher numbers of arthropods were sampled in summer than winter. Dead trees had between 0.69-0.84 as many arthropods alighting on them as live trees (intercept trap data), and between 0.36-1.00 as many arthropods living on them as live trees (bark trap data). Levels of many orders on dead and live trees were not significantly different from each other. The variety of beetle species was much lower on dead than live species, although species composition was different. It is concluded that retention of dead trees provides habitat for arthropods, enhances the conservation of biodiversity, and provides additional feeding opportunities for insectivorous vertebrates, that would not be available if they were removed.

Reliability of plants & various groups of animals as indicators of environmental condition: a cross taxon analysis

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A survey of rehabilitated mineral sand mines and reference heath areas at Eneabba, Western Australia was conducted in order to compare the relative performance of plants, birds, terrestrial vertebrates, and various orders of arthropods as indicators of environmental conditions. Using both PATN, and the 2stage application in the PRIMER5 multivariate package, ants and beetles performed almost as well as plants as indicators of overall environmental conditions. Terrestrial vertebrates and springtails performed acceptably well, but trends in birds and spiders were more closely allied to trends in other taxa than they were to the overall community. A critique of the performance of each taxon is presented.

Conservation status of the mound-building red wood ant *Formica lugubris* in Ireland

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Formica ants form a substantial component in coniferous forest ecosystems in the northern hemisphere. These locally dominant wood ant species are included in the global red list of IUCN as a result of habitat loss and degradation. In Ireland, the distribution and population size of *Formica lugubris* has declined rapidly in recent decades. Only two small isolated populations remain and the total number of known nests is less than 50 (compared to 200 during the 1970s). For conservation purposes, more information about the species status is needed. We use genetic methods, microsatellites and SSCP, to examine sociality, genetic variability and structure of the populations, and to clarify the origin of *F. lugubris* in Ireland. The genetic variation is extremely low, but there exists only little inbreeding depression measured as the proportion of diploid males. Both populations are monogynous having normally one queen per nest, whereas populations in England have been shown to be polygynous. Thus, Irish *F. lugubris* populations differ from those in England, indicating an even stronger need for conservation. In addition, *F. lugubris* needs urgent conservation as the effective population size is really low and very little genetic variation is left in the remaining populations threatened by forestry.

Why do ants distribute myrmecochorous diaspores? New insights to an old question

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Ant-dispersed diaspores usually bear a protein and lipid-rich appendage, the elaiosome. The attractiveness of elaiosomes for ants has been repeatedly demonstrated. The nutritional composition of elaiosomes, however, has received little attention so far. In our study, elaiosomes and the respective seeds of 15 Central European ant-dispersed diaspores were chemically investigated in regard to the possible nutritional requirements of ants. The results suggest that elaiosomes are adapted for the needs of ants. Investigating the distribution of *Corydalis cava* elaiosomes between larvae and working caste of laboratory colonies of *Myrmica rubra* we found them to be mainly consumed by larvae and not by workers. As Oberrath and Böhning-Gaese (2002) could show myrmecochorous plants fruited on average 7.1 weeks earlier than those with other dispersal modes. So both, the highly nutritive elaiosomes and timing the fruiting peak with the seasonal peak of larval abundance are evolutionary adaptations of plants towards myrmecochory. In view of these results the hypothesis, that 1,2 diolein, a diacylglycerol of oleic acid act as trigger for seed dispersal, was examined once more. In a series of bioessays we tried to clarify whether ants prefer single substances over mixtures with nutritionally valuable components. The results give reason for a new discussion of this hypothesis.

The role of hybridization & genetics in landscape dominant sagebrushes (*Artemisia*)

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The subgenus *Tridentatae* of *Artemisia* (Western North America sagebrush) is composed of a group of about a dozen species. Several species including iconic big sagebrush (*A, tridentata*) encompass subspecific entities. The *Tridentatae* as a whole are closely related. Polyploidy is a reoccurring theme. Studies using hybridization, genome analysis, allozymes, and DNA-molecular genetics on individual populations, species, and higher taxonomic ranks have been useful in clarifying systematics. Classical morphological (alpha) taxonomy has been partially confirmed. Molecular genetics approaches using RAPD, AFLP, ITS, and *trn*L have provided new insights on the limits of *Artemisia* and of *Tridentatae*. The genetic structure of the *Tridentatae* as a whole suggests a group differentiated as a result of strong ectotypic selection over a background of widespread hybridization—chloroplast and ribosomal DNA patterns are not taxon specific. However, some AFLP markers show high subspecific fidelity within *A. tridentata*. Local gene flow and strong evidence for population adaptation has been provided by a series of studies in hybrid zones. Insects have variable responses to parental and hybrid sagebrush populations both in natural hybrid zones and manipulated parental-hybrid sagebrush plantations but the hybrids do not appear to be a sink for herbivorous insects or to have lower herbivore loads.

Status evaluation principles for insects in New Zealand

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An outline is provided of the principles underlying the system utilised by the New Zealand Department of Conservation (DOC) to assign threat rankings to species. A brief history of previous status evaluation systems used by DOC and background into the development of the current system is covered. Advantages, assumptions and drawbacks of the current system are discussed. A synopsis of how the rankings are integrated into departmental processes and their influence on the management of insect species is provided.

Tracking large rare weta (Orthoptera, Anostostmatidae)

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The largest of the giant weta, *Deinacrida heteracantha*, were formerly widespread in northern forests, but now survive only on Little Barrier Island (New Zealand). Rugged topography of the island and arboreal habits of the weta make it difficult to assess their current status, but it is a concern that they are no longer seen in the low coastal shrubs where they were described as 'easy to find' in the 1960s (Richards 73). Surveillance of individual weta using radiotransmitters for up to 25 days of monitoring indicates some aspects of habitat use, with implications for their continued co-existence with kiore *(Rattus exulans)* and for future monitoring.

Accuracy of species richness estimates obtained from incomplete inventories: lessons from a benchmark dataset on tropical ants

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Estimating species richness of insect communities is notoriously difficult especially in the tropics where unrecorded species continue to appear in the samples even after years of effort. Methods allowing to extrapolate accurately the species richness (ESR) for larger sampling efforts or the total species richness in the assemblage (TSR) are highly desirable to improve the cost effectiveness of biodiversity inventories. Here, we tested the performance of 1 parametric, 5 non-parametric and 15 curve-fitting extrapolation methods by calculating their bias and precision against a large benchmark dataset, a near complete inventory of a tropical ant assemblage. For low levels of sampling effort (before the species accumulation curve started to level off), beta-P and Soberon & Llorente models allowed extrapolations of species richness for up to 10-fold increases in sampling effort with little bias and reasonable accuracy. There is, however, a minimum sample size below which, little information can be gained, regardless of extrapolation methods. For higher levels of sampling effort the Stout & Vandermeer and the Weibull models, yielded stable estimates of TSR, independent of sample size. Overall, curve-fitting extrapolation methods appeared as a powerful alternative to the popular non-parametric methods.

Discriminatory power of arthropod surveys conducted at family level for monitoring disturbance in tropical forests

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Arthropods were surveyed by local parataxonomists at 12 sites of increasing anthropogenic disturbance (old and young secondary forests, savannah and cultivated gardens) at Gamba, Gabon. Overall 429,541 arthropods were collected in the 38 sampling events (4,712 samples in total) carried out over one year with four sampling techniques: yellow pan, pitfall, flight interception and Malaise traps. Here, I report on the discriminatory power of family level data to monitor disturbance in tropical forests as an alternative to using a single taxon treated at species level. The families most useful to classifying sites along the disturbance gradient were identified on the basis of their indicator value index for each sampling method separately. Discriminant analyses were then performed to select the most effective predictor set of families. The discriminant functions derived from the analyses of the whole dataset were subsequently tested on subsets of the original dataset to determine 1) the minimum sample size recommended for efficient monitoring, 2) the most frequent sources of error and 3) the most accurate sampling method. Overall, this study shows that analysing arthropod surveys at family level can be an effective alternative to a single taxon approach for monitoring disturbance in tropical forests

Current status of the Odonata of Bhutan: a checklist

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Lieftinck (1977) has reported only six species of dragonflies from Bhutan. Tsuda (1991) in his distribution list of world odonata has listed 9 species of dragonflies from Bhutan. Mitra (2002) in his recent most work found out 18 new records from Bhutan. In the present study an up-to-date checklist of 34 species and subspecies of dragonflies known from Bhutan has been made of which seven are new records from Bhutan.

Guanophilic arthropod ecology & seasonal spatial distribution in a bat maternity cave

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Australia's most diverse guanophilic arthropod community from Bat Cave, Naracoorte consists of over 30 species of insects, arachnids and crustaceans. Highest species abundances occur during summer when the maternal chamber is occupied by some 30,000 southern bent-wing bats that use the domed chamber for birthing. During summer deposition of fresh guano and urine is greatest in the chamber's centre, under the main bat roost, resulting in dramatic increases in pH, while remaining areas exhibit lower pH. During winter bats vacate the cave to enter torpor in cooler caves in the region, resulting in a cessation of guano deposition and consequently lower, often acidic conditions (pH <5.0). High abundances of guanivorous mites and predatory histerid beetles occur on guano piles during summer corresponding with increased guano deposition rates and high pH (>8). Older more acidic guano in winter enables large population increases of anobiid and other fungal feeding beetles. These species dominate guano piles in the central area. Generalist predatory species including carabid beetles and pseudoscorpions are ubiquitous in the maternal chamber throughout the year and show no preference for position of guano within the chamber. This highly specialised ecosystem provides an invaluable study site for subterranean ecology and cave biogeography in southern Australia.

Body size variation in Australian Coleoptera: comparing canopy assemblages from remnant & plantation forests

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Body size of insects is related to many ecological traits including diet, reproduction and dispersal. Comparing body size distributions of communities from different habitats informs us of ecological differences over and above that which can be learned from species composition alone. We collected, identified and measured body lengths of 273 beetle species from the canopy of remnant *Eucalyptus marginata* forest and *Eucalyptus globulus* plantation forest from the same region in southwestern Australia. Four survey events were conducted (spring and autumn over 2 yrs) from 8 remnant forest and 15 plantation sites across 4 study areas. We used insecticidal knockdown from canopies of 26 remnant *Eucalyptus marginata* and 53 plantation *Eucalyptus globulus* trees. The mean body size of species (ignoring abundance) was larger in the plantation habitat (t-test, 144 df, P=0.002). The mean size of individuals (ignoring species) was also larger in the plantation habitat (t-test, 5000 df, P<0.001). However, the few most abundant species were not consistently larger than their near-relatives (within-family). Smaller species from both habitats were found to occupy more sites than large species. This relationship between body size and number of sites occupied was over and above the effect of abundance (logistic regressions, 2 independent variables, P<0.001).

The effect of climate change on insect ranges: dynamic profile models

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The consequences of climate change depend on the species' ability to track it. I model the effect of a moving North-South climate profile on the latitudinal range of an insect metapopulation. It is assumed that climate affects one or more of the metapopulation parameters (amount of habitat that is regionally available, local extinction rate and colonization rate). When the ranges shift in response to the climate, tracking delays may develop on both the advancing and the retreating edge of a range. The difference between these two delays determines the effect on range size. It appeared that a moving climate lowers regional population size. However, ranges may become larger when populations on the retreating edge take a long time to disappear. Remarkably, global population size may increase. Species not able to track the climate fast enough go extinct because their retreating edge moves faster than, and eventually collides with, the advancing edge. Results depend very much on which of the metapopulation parameters climate actually acts on, especially concerning the retreating edge. Tracking ability increases with range size, amount of habitat, dispersal rate and dispersal distance, and climate change greatly increases vulnerability to habitat destruction and dispersal impairment.

Soil & litter arthropod assemblages as indicators of rainforest restoration

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The devastating impacts of forest clearance are widely recognized, and considerable efforts have been employed to restore cleared areas. However, little is known about the re-assembly of soil and litter fauna in replanted areas, or how assemblages may be affected by different restoration practices. The present study investigates factors that influence the diversity of soil and litter dwelling arthropods in rainforest restoration plantings. Factors include: distance from remnant rainforests; inoculation; litter quality and quantity; and the efficacy of timber plantations versus ecological restoration. A field experiment is in progress to test these factors. The first stage is to select suitable indicator groups by means of a baseline survey. A set of 24 replicate sites was located in pasture and rainforest on the Maleny plateau, in the Sunshine Coast hinterland of eastern Australia. Arthropods were sampled using pitfall traps and Tullgren funnels. Various arthropod taxa are used in the present study, with particular emphasis on ants (Hymenoptera: Formicidae). The usefulness of different taxonomic levels as indicators of rainforest restoration will be compared.

Inter-mutualism conflict: do ant bodyguards deter pollinators?

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We tested the hypothesis that plant bodyguards (ants) also deter plant pollinators, and explore mechanisms by which a plant bearing extrafloral nectaries (Ferocactus wislizeni, Cactaceae) may limit inter-mutualism conflicts between its pollinators (solitary bees) and a diverse guild of ant bodyguards. No-choice trials demonstrated that floral tissue can deter ants, although that deterrence differed among four ant species. Field inspections of >1000 flowers on plants with different ant associates demonstrated that ant occupation of flowers (the product of % flowers occupied and mean number of ants within an occupied flower) increased in the order predicted by the behavioral trials, and the percentage of flowers occupied by bees decreased in that order. Ant identity also influenced maternal plant reproductive function. Seed mass per fruit was significantly lower on plants tended by Solenopsis xyloni, the ant least deterred by floral tissue, relative to fruits on plants tended by the other three species. That ant exhibited the greatest incidence of flower occupation, the greatest aggressiveness towards other arthropods, and pollinators visited plants tended by that ant least. Inter-mutualism conflicts may be particularly common in facultative, generalized systems, particularly where there is variation in partner quality and in the relative importance of the mutualisms.

Improving conservation focus: refining status evaluation for threatened insect species

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Deciding rational priorities from lists of protected species to receive limited support resources has implicit reference to both absolute need and ranking. Both of these are often somewhat subjective decisions, but purport to reflect imminence or likelihood of extinction. Criteria used to determine conservation need for invertebrates are commonly derived from those designed for vertebrates, and contain reference to quantitative data on extinction thresholds and population sizes of little practical relevance, and which are usually not available. Shortcomings of current schemes for setting such priorities are reviewed, and the distinction between 'rarity' and 'threat' emphasised. A scheme designed to evaluate conservation status of Australian butterflies, based on severity of threat, is outlined and its potential wider applications discussed. Such schemes lead toward more credible and workable priorities for conservation need and may do much to overcome problems of the current listing processes, so leading to better focus and allocation of the limited support available for insect species conservation.

Does a wide host plant range promote speciation?

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Phytophagous insects represent a large proportion of global biodiversity, but how and why this diversity has evolved is not clear. The butterflies are an important model group for investigation of this pattern. We make use of recent advances in plant and butterfly systematics to trace the evolution of host plant utilization at several taxonomical levels in the family Nymphalidae. A consistent pattern was found that butterfly taxa with a more diverse utilization of host plants also are more diverse in terms of number of species. We discuss this pattern in terms of Ehrlich & Raven's coevolutionary scenario of "escape and radiate" and West-Eberhard's theory of plasticity as a driver of speciation.

Tracing the great Australian weevil radiations

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The Australian weevil fauna of ca. 4200 described species is characterised by a small number of highly diverse groups that are unique to Australia or not numerous elsewhere. These taxa can be categorised as terricolous (Amycterini, Entimini), phytocolous (Belinae, Gonipterini, Cryptoplini, Storeini) or lignicolous ("Cryptorhynchinae"). All are indicated to have undergone significant radiations; however, understanding the nature and drivers of their radiations is largely impeded by the poorly known taxonomy and phylogeny of the groups. In the phytocolous Belinae, whose taxonomy and phylogeny have recently been studied, the radiation can be traced on a phylogram as being driven by a host shift from conifers to acacias. Radiations in other phytocolous taxa similarly appear to track the diversification of Australian acacias (Storeini) and Myrtaceae (Gonipterini, Cryptoplini). When the phylogeny is unknown, radiations may be estimated from the structure of the diversity. In the terricolous Amycterini and Entimini, diversity profiles suggest the existence of radiations of different ages, origins and drivers. Diversity structure appears largely unaffected by the state of taxonomy of a group: in Amycterini, revisions in 1865-1866, 1909-1923 and 1993 resulted in changed numbers of genera and species but not diversity structure.

Collapse of an island insect fauna due to invasion by an exotic lizard, *Anolis carolinensis*

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The Ogasawara (Bonin) islands, located between mainland Japan and the Marianas, have a unique fauna like many other Oceanic islands in the Pacific. The insect fauna is not so rich but is highly endemic – so much so that a species of butterfly, a cicada, four dragonflies and damselflies, a bee, and one jewel beetle are protected as national monuments. Recently, however, their populations have been declining and have become endangered. These declines were thought to have been caused by previous human development of the islands, but this does not explain why some groups are not declining. Butterflies, a cicada, bees, dragonflies, damselflies, and some groups of beetles are declining, while moths, water striders, and other groups of beetles are not. We observed that the species in decline were diurnal and that this decline had begun after the invasion of a diurnal lizard, *Anolis carolinensis*, the green anole, which was originally distributed in southern North America, but has been introduced to a number of Pacific islands. Field and indoor experiments showed that the green anole is an effective insect predator and is the likely cause of the declines

Forest edges: regions of high or low diversity in plantations?

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Managing plantations for diversity as well as production can be enhanced by taking into account local and landscape level structural heterogeneity. Forest edges contribute to such heterogeneity and may have positive and/or negative effects on diversity. Although edge-effects have been demonstrated to occur for microclimate, vegetation assemblages and some vertebrates, there are few data for arboreal arthropods communities. This paper presents data on edge-effects for canopy arthropods in UK coniferous plantations. Arthropods were collected using pyrethrum knockdown from a range of patch sizes and tree species. Groups such as the Acarina, Coleoptera and Collembola showed strong responses to edge proximity with some species exhibiting marked changes in density or clear habitat preferences. Responses of canopy communities varied with tree species, for example, in patches of the introduced species Picea sitchensis, there were elevated levels of herbivores and predators at the edge whereas in Pinus sylvestris (native to Caledonia in Scotland) the core habitat supported more diverse arthropod communities with greater numbers of woodland specialists. Recommendations about management strategies for increased biodiversity are discussed.

Impacts of climate change on insect communities

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Observed changes in natural systems, largely over the past century, indicate a clear global climate change signal. Even in the face of apparently dominating forces, such as direct, human-iven habitat destruction and alteration, this climate fingerprint implicates global climate change as a new and important driving force on wild plants and animals. Analyses of climate and of biotic change indicate that on a global scale, climate change has affected about half of the wild species studied (Parmesan & Yohe, 2003). One of the most prominent responses has been poleward and upward shifts in species' ranges, apparently tracking climatic shifts. The implications of shifting species' ranges for maintenance of species' interactions and conservation of local diversity will be discussed.

Ants of Kruger National Park, South Africa: diversity, determinants & conservation implications

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Epigaeic ant diversity was surveyed across a range of savanna habitats in Kruger National Park, South Africa, in order to compare diversity between habitats, and investigate the relative importance of local scale processes responsible for maintaining this diversity. A total of 204 ant species were recorded in the park. No invasive or alien species were found in the park. Species richness was greatest in the highest rainfall area (Pretoriuskop), and lowest in lower rainfall areas. Although fire is generally considered an important disturbance in Southern African savannas, burning was not found to significantly affect ant diversity across most of the park. Higher rainfall areas where ant community composition was significantly affected by fire regime, appear to be an exception. While habitat structure did not affect ant body size, ant community composition differed between savanna habitat types. Competition was found to play an important role in determining ant diversity at baits, although how this is reflected at the assemblage level still requires further investigation. The findings have implications for conservation management in Kruger National Park, and will assist with the choice of sites for long-term monitoring.

Caught in the web: assessing spider assemblages as ecological indicators in agroecosystems

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Spiders are important ecological indicators that reveal the impacts of environmental change and more importantly management decisions in agroecosystems. Plant and vertebrate species diversity have been frequently used as ecological indicators in natural environments however in agroecosystems these taxa are less useful. Spiders on the other hand satisfy all the requirements of a good ecological indicator in that they are relatively diverse and abundant in most agricultural regions of the world, are easily sampled, and respond rapidly and predictably to their environment in a way that reflects ecological change. Their trophic position means they may have major impact on other invertebrates and are functionally significant as predators of agricultural pests. Previous studies in agroecosystems have shown that spider species richness decreases as farm management intensity increases. The potential use of spider communities to monitor tillage regimes, insecticide usage, and the success of area-wide integrated pest management programs is discussed.

The relative abundance of leaf domatia & extrafloral nectaries in South Florida trees & plant communities: temperate versus tropical comparisons

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Southern Florida is both the southern limit of many temperate trees and the northern limit of many tropical trees. The leaves of most of the trees in the region (122 species) were examined for the presence of leaf domatia and extrafloral nectaries, structures promoting potential plant protection mutualisms with beneficial mites and ants. Leaf domatia were found in 7/21 (33%) trees of temperate origin compared to 15/101 (15%) of the trees of tropical origin. Extrafloral nectaries were found in 2/21 (10%) of the temperate trees compared to 23/101 (23%) of the tropical trees. Plant communities comprised principally of temperate trees (Scrub, Cypress and Everglades Tree Islands) should have relatively high occurrence of leaf domatia based mutualism but low levels of extrafloral nectary based mutualism, with the reverse being true for plant communities comprised primarily of tropical trees (Tropical Hardwood Hammock and Pine Rockland). Tropical Mangrove, however, has similar levels of occurrence of leaf domatia and extrafloral nectary bearing trees. These results agree with past studies showing a greater abundance of extrafloral nectaries in tropical plants, and support findings indicating that leaf domatia are especially common in temperate trees, but suggest clearer tropical vs. temperate delineations in these protective mutualisms.

Evolutionary constraints & adaptation in psyllids

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Evolutionary constraints may both promote and restrict adaptation. An obvious way in which specialist phytophagous insects "break" evolutionary constraints is in switching to different host plants. The diversification of plant feeding insects depends on the ability to colonize novel plant groups, but also to specialize on these groups. In many cases, specialization has promoted spectacular radiations in insects, promoting adaptation and diversification. In Australia, there are several examples where the scale of diversification in the host plant appears to have promoted diversification in the insects (e.g. psyllid diversification on Myrtaceae, Fabaceae and Casuarinaceae). The diversification of a plant lineage may influence the development of insect specialization in a number of ways. Joint analysis of associated plant and insect groups is one way to answer some of the questions about the role of evolutionary constraints and adaptation.

Lessons from the mysterious east: the tectonic and biogeographic history of the East Papua composite terrane

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The tectonics of the eastern Australian Plate margin from the Cretaceous onward were dominated by an initial episode of rifting during which continental slivers moved northeastward into the Pacific, followed by eastward-dipping subduction in the Eocene during which a set of island arcs converged upon the margin in a westward direction. Following subduction reversal to a westward dip in the Oligocene, the predominant direction of arc migration became eastward, the pattern that prevails today. Thus an initial outward rafting of old Australian biota comingled with an inward rafting of more heterogenously derived arc biotas which originated in both Asian and oceanic settings; the results of this biotic interchange are now being rafted eastward into the Pacific at large. In eastern New Guinea, the East Papua Composite Terrane, or EPCT, was caught between these dichotomous tectonic regimes for at least 30 million years, and provides an instructive case history in regard to biotic derivation and fragmentation. The EPCT developed a distinctive endemic biota that still imparts a strong regional signature, and provides useful independent evidence for evaluating models of tectonic evolution in the Solomon Sea region.

Codiversification in an ant–plant mutualism: stem texture & the evolution of host use in *Crematogaster* inhabitants of *Macaranga* in Southeast Asia.

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We investigate the evolution of host association in a cryptic complex of mutualistic *Crematogaster* ants inhabiting *Macaranga* trees in Southeast Asia. We use Cytochrome Oxidase I (COI) to reconstruct phylogenetic relationships in a comprehensive sampling of *Crematogaster* from *Macaranga*. Using a published *Macaranga* phylogeny, we test whether the ants and plants have cospeciated. The COI phylogeny reveals ten well-supported lineages, and an absence of cospeciation. Host shifts, however, have been constrained by stem traits that are themselves correlated with *Macaranga* phylogeny. Despite the absence of cospeciation, a correlated succession of stem texture in the ant and plant phylogenies suggests that *Decacrema* and *Pachystemon* have diversified in association, or codiversified. Their confinement to the aseasonal wet climate zone of western Malesia suggests myrmecophytic *Macaranga* are no older than the wet forest community in Southeast Asia, estimated to be ~20 million years (MY) old. Applying a COI divergence rate of 1.5% per MY to a rate-smoothed Bayesian chronogram of the ants, the *Decacrema* from *Macaranga* are inferred to be at least ~12 MY old, suggesting that the evolutionary histories of *Decacrema* and their *Pachystemon* hosts have been contemporaneous since the mid-Miocene. The evolution of myrmecophytism enabled *Macaranga* to radiate into enemy-free space, while the ants' diversification has been shaped by stem traits, host specialization and geographic factors.

Impact of anthropogenic disturbances on litter insect communities in the heavily used forests of Western Ghats (India)

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"In most parts of the Western Ghats, stand simplification due to human use of forests is the rule rather than exception. In Sringeri Taluk of central Western Ghats, [Karnataka, India] there is a traditional practice of leaving small fragments of forests uncultivated near the farm lands, for the purpose of green manure and fuel wood and are called *Soppinabettas*. Due to over exploitation the diversity of these *Soppinabetts* are depleting very fast and are being converted to less diverse scrub jungles. On the other hand, state forest policy led to the complete replacement of "natural" or "degraded" forests with monocultural *Acacia auricularis* plantations thus further simplifying the vegetational complexity. A study was conducted to compare litter insect diversity in relatively less disturbed natural forest patches with that in well-managed *soppinabettas*, degraded *soppinabettas*, and acacia plantations. Using satellite imagery and GIS techniques, five sites containing each of these four land-use types were identified.. Insects sampling was carried out across three seasons using pitfall traps and Berlese-Tullgren funnels. Data on environmental variables, vegetation structure and litter density were also collected. The results show that species richness is maximized in the well-managed *soppinabettas* while acacia plantations are the least diverse. Insect abundance is maximised at 'intermediate levels of disturbance' and increased frequency of disturbance limit diversity to those species can tolerate stress due to disturbances.
"

Here today, gone tomorrow: selecting appropriate bioindicators for temporary habitats

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Temporary habitats, such as floodplains and ephemeral ponds, are important on a worldwide scale for many rare and threatened invertebrates. The conservation status of these systems is difficult to assess and monitor because of their temporal instability. Turloughs are temporary water bodies that occur in the west of Ireland. As unique habitats, turloughs are designated for nature conservation under the European Union Habitats Directive. Ireland is therefore obliged to designate sites for conservation and monitor their habitat quality. There are a number of questions that this paper addresses. Firstly, are the conservation agencies taking appropriate account of all aspects of turlough biodiversity? Should both phases of the habitat be monitored? Or can terrestrial invertebrates be used as bioindicators for the aquatic invertebrates and vice-versa? This paper considers invertebrate communities of both the dry and the wet phases of turloughs. There were a number of similarities between the terrestrial and aquatic assemblages. However, the major environmental factors influencing the groups differed, with some sites being noteworthy for their terrestrial fauna, but not their aquatic fauna, and vice versa. This work sheds new light on the conservation and management of temporary habitats.

The adventure of few and gain of many: adaptive radiation of spiders & planthoppers on remote islands of the Pacific

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Remote islands have been heralded as "natural laboratories", with spectacular cases of adaptive radiation. How is the diversification achieved? To elucidate commonalities underlying patterns of adaptive radiation we have studied (1) spiders in the genus *Tetragnatha* which are generalist predators; and (2) planthoppers in the genus *Nesosydne* which are specialist herbivores. Comparing three archipelagoes of differing isolation across the Pacific we have shown that both genera have undergone adaptive radiation within each archipelago, although the lineages within each group are only distantly related. Focusing on the Hawaiian Islands, where diversification is most prolific, we show that lineages have generally progressed down the island chain from older to younger islands, a common pattern for arthropod radiations. For the spiders, species on any one island are generally closely related to others on the same island, and the same set of ecological forms has evolved repeatedly on different islands. The planthoppers show a similar pattern of within-island proliferation, though diversification is associated with plant diversity, some lineages tending to diversify in concert with their host (host tracking), while others diversify through host switching. Comparison of diversification in the spiders and planthoppers allows us to infer general mechanisms underlying adaptive radiation.

Alien invasive species in Galapagos: challenges for the conservation of insect diversity

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The conservation of the approx. 2000 indigenous Galapagos insect species is being threatened by the presence of introduced species including almost 500 insect, 600 plant and 15 vertebrate species. Population decline and even extirpation of endemic Galapagos insects are directly related to predation by invasive species such as *Polistes* wasps and fire ants, and indirectly affected by herbivores such as goats and the cottony cushion scale, and invasive plants. To reduce the risk of new incursions, an inspection and quarantine system for the Galapagos Islands was set up by a multi-institutional task force. At the same time, the Charles Darwin Foundation has developed an integral strategy to conserve the unique Galapagos insect fauna. An ecological monitoring system has been developed that acts as an "early warning system" for new species arrivals and provides important data for the analysis of ongoing mitigation and restoration actions. A model is being developed to predict the invasiveness of potential quarantine insect pests and insect species that have already been introduced in order to prioritize management actions. Several successful eradication and control programs have been executed to deal with invasive species such as the little fire ant, the cottony cushion scale, goats and invasive plants.

Specialist seed insects can affect the conservation of endangered conifer species

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Most insects exploiting conifer seed cones are highly host- specific. Several ecological adaptations, especially prolonged diapause, allow these insects to cope partially with the annual variations usually observed in host abundance. Long-term, standardized surveys were carried out in Europe and China in order to assess subsequent insect impact on the potential of natural regeneration of conifers. In widespread species (e.g., larches, *Larix* spp.), insect impact dramatically decreased the potential seed yield but a sufficient natural regeneration occured when unforeseeable climatic events (e.g. late frosts) perturbed the natural synchrony between insect populations and cone abundance. Contrasted results were obtained for endangered conifers. In incense- juniper (*Juniperus thurifera*), a relict species with a very scattered range, low numbers of initial seeds per cone in Morocco and France resulted in a quasi- absence of viable seeds following insect attack whereas higher seed numbers allowed an important natural regeneration in Spain. In China, intensive forest exploitation limited the range of *Keteleeria xerophylla* and *Pseudotsuga sinensis* to very small, isolated plots where up to 100% of the seeds were annually destroyed by insects. Insect control seems the only way to prevent species disappearance.

Invasive alien plants & insect diversity conservation

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Invasive alien plants (IAPs) are among the most threatening of processes to insect diversity. The magnitude of the impact is uncertain, but is large and increasing. The impacts of IAPs at the local scale on various indigenous insect taxa can be negative, neutral and, in some cases, positive. Similarly, the planting of alien trees for timber can be variable depending on the types of alien tree and indigenous insect. Using examples from Africa and the Western Indian Ocean islands, a range of situations and outcomes will be given in the search for general patterns. These findings are then used as a baseline for developing a strategy for ameliorating the threats and searching for opportunities to optimally manage to secure the future of insect diversity.

Habitat fragmentation and connectivity changes hymenopteran parasitoid community composition but not overall diversity

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Higher trophic levels (parasitoids and predators) might be more sensitive to habitat fragment area and isolation than species at the lower trophic levels. We investigated the hymenopteran parasitoid communities in native-forest fragments (small-isolated, small-connected, large-isolated, large-connected) ranging from 473 ha to 10685 ha over a period of two years from Wellington in New Zealand. MDS and analysis of similarities (ANOSIM) showed a significant difference in species composition between fragments (R 0.413, p=.001). However, there was no effect of fragment size or connectivity on the overall diversity (Shannon's H') of parasitoids. Species composition changed between different sized fragments and ratios of exotic: endemic taxa changed between various fragments, though not necessarily according to size or connectivity. In a case study, we examined the abundance of the kawakawa moth (*Cleora scriptaria*) and its parasitoid fauna, primarily consisting of an endemic and exotic braconid wasp species between fragments. Fragment size and connectivity did not influence the abundance of the moth larvae, but resulted in lower parasitism rates by the exotic braconid wasp. These results demonstrate how community composition can significantly change while overall levels of biodiversity remain similar. Thus, attempting to make decisions on the conservation value of individual habitat fragments becomes complicated.

Effects of reduced prey choice caused by GM plants on a predatory insect

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Bt plants are highly toxic to certain pest species. Predatory insects may be affected by the reduced abundance of target pests if sufficient alternative prey is not available. This paper will present experiments investigating such indirect effects under laboratory conditions. Lacewing (*Chrysoperla carnea*) larvae were released into large cages containing Bt and wildtype plants previously infested with a constant number of diamondback moth (*Plutella xylostella*) eggs but different numbers of aphids (*Myzus persicae*). No *P. xylostella* larvae survived on Bt plants but aphid populations reached high densities on both plant types. Lacewings recovered from Bt plants at the end of the experiment did not differ in weight from ones recovered from wildtype plants. At the highest initial aphid densities there was also no difference in the proportion of lacewings surviving on the two plant types. Significantly fewer lacewings were recovered from Bt plants than from wildtype plants as initial aphid numbers were reduced. Interestingly, however, numbers of *C. carnea* recovered from wildtype plants were only significantly correlated with numbers of surviving *P. xylostella* at the highest aphid densities. Possible roles of escape behaviour of *P. xylostella* as well as cannibalism between *C. carnea* larvae in these interactions will be discussed.

Community conservation of threatened flagship species: with reference to recovery of the Richmond birdwing butterfly

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Community participation has been effective for recovering threatened butterfly species and is a valuable tool for invertebrate education in Australia. Flagship species of conservation concern have been actively managed in South Australia, Victoria, New South Wales and Queensland. Community groups have focussed on protecting habitats and by protecting specific food plants at the breeding sites. In Queensland and New South Wales, a Project on the Richmond birdwing butterfly, *Ornithoptera richmondia* (Gray), started by a CSIRO entomologist and a national parks ranger, linked with educational and environmental personnel, to conserve the butterfly. The Project commenced in 1989, when the lowland butterfly food plant, the vine *Pararistolochia praevenosa*, was cultivated by a plant nursery for distribution to schools and community groups. An Officer from CSIROs Double Helix Science Club later coordinated activities, and by 1998, more than 32,000 vines had been distributed from the nursery. Support from CSIRO, Brisbane's Southbank Corporation, Bayer Australia and a Threatened Species Network Community grant, enabled wider community (8 groups) and participation in workshops. Progress has been achieved after more than 10 years, by providing ecological guidance to the community (>2000 members) and schools (>200 involved) towards conserving the butterfly, and management and restoration of its habitats.

Evolutionary origins of cave cockroaches in the genus *Paratemnopteryx* Saussure (Blattodea: Blattellidae), Australia

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For over 150 years scientists have debated the origins of cave dwelling organisms. To investigate the origin of cavernicolous fauna in North Queensland, we selected the cockroach genus *Paratemnopteryx* Saussure (Blattaria, Blattellidae) to test alternative hypotheses for the evolution of cave dwelling species within the genus. Our initial hypothesis was that some surface dwelling species within this genus gave rise to cavernicolous species as a result of increasing aridity in Australia during the late Cenozoic, based on Vandel's 1965 Pleistocene-effect model for the origin of cave organisms in temperate regions. To test this, we used a combined morphological (30 characters) and molecular (ITS2 and COII) approach. Results indicated that cave colonisation events occurred independently on at least three occasions, once in Western Australia and twice in North Queensland. Evidence from species distributions, the structure of the phylogenetic trees, and the possible timings of cave colonisation events (calculated from the maximum likelihood analysis), support the Pleistocene-effect model for the evolutionary origin of cave cockroaches in the genus *Paratemnopteryx*. Increasing and spreading aridity over much of Australia in the late Cenozoic may have provided strong selective pressures for surface dwelling species to colonise caves in search of moisture refugia.

Are invertebrate focal taxa fulfilling their promise?

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Pervasive scarcities of data on the distribution, taxonomy, and population status of invertebrates have seriously impeded the inclusion of invertebrate diversity in the conservation process. For nearly two decades, the development of invertebrate focal taxa has been suggested as a means to pry conservation-relevant information out of this hyper-diverse, poorly known invertebrate realm.. A review of our progress toward establishing such focal taxa reveals decidedly mixed success. While there have been huneds of studies debating the criteria for selecting focal taxa and detailing the effects of anthropogenic disturbances and ecological changes on dozens of different invertebrate taxa, they have resulted in a fragmented and often incomparable dataset. As a result, virtually none of the many proposed invertebrate focal taxa have been universally embraced, though a slate of promising candidates has been identified. However, we have developed a clearer understanding of the informational, methodological, and taxonomic infrastructure needed to support fully functioning focal taxa, and the coordinated effort necessary to produce it. Rapidly improving information technologies and more focused efforts by collaborative networks of taxonomists, ecologists, and conservation biologists have the potential to rapidly develop a suite of invertebrate focal taxa with utility in a variety of conservation contexts.

Does biomonitoring guard against loss of rare, threatened or endemic species?

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Extensive biomonitoring programs are being recommended and established worldwide with the intention of protecting various jurisdictions from loss of biodiversity. Despite the fact that arthropods account for a huge share of biodiversity and are demonstrably at risk from large-scale anthropogenic impacts on landscapes, proposed biomonitoring schemes frequently ignore well understood aspects of arthropod assemblages. For example, detection of many arthropod species can be highly seasonal, and thus, short pre-planned visits do not give reliable data about the presence of a species at a particular site. Most importantly, as will be illustrated with data sets from many taxa, our most 'threatened' species are also our rarest and their loss will be undetected by any but the most thorough studies. Simplisitc monitoring studies, planned without reference sites, forgo strong cause and effect inferences about the causes and, hence, proper response to species declines. Rather than the 'half-baked' apple pie and motherhood of senseless monitoring, we recommend careful consideration of objectives and development of more rigorous efforts to ascertain effects of anthropogenic disturbance on vulnerable elements of invertebrate biodiversity.

Do invertebrates use willows? The influence of landscape matrix on the impact of an invasive weed

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Native animals, particularly invertebrates, often respond negatively to invasive weeds because they are often associated with resources that are plant specific. However, the surrounding landscape is also likely to have a strong influence on native fauna and their use of resources. Crack willow (*Salix fragilis*) has been used extensively for erosion and flood control in New Zealand and is now an invasive weed, particularly in riparian zones. This study aimed to quantify the use of willow trees by invertebrates and to determine whether willow use by invertebrates is dependent on landscape matrix. We examined invertebrate communities associated with willow or native kanuka (*Kunzea ericoides*) trees in riparian zones surrounded by either native forest or pasture. Invertebrates responded strongly at the level of tree species; the larger landscape matrix appeared to have little influence on invertebrate abundance, richness and composition. Lower invertebrate abundance and richness were associated with willow trees. However, there were beetle species-level responses to both tree species and landscape type. Kanuka trees surrounded by pasture had similar invertebrate compositions as kanuka trees within intact forest, indicating that small remnant patches of kanuka are valuable invertebrate habitat.

Moth beta-diversity patterns in Papua New Guinean rainforest

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Local and large-scale patterns of beta-diversity in moth assemblages have been examined in forest habitats in Papua New Guinea, in a project funded by the UK Darwin Initiative. Moths were surveyed quantitatively by light-trapping and identified to morpho-species. Undisturbed primary forest sites and areas that have been re-vegetated as environmental remediation after open-pit gold/silver mining were compared on Misima Island, Milne Bay Province. Standard community analyses indicated that natural forest and re-vegetated areas were remarkably similar in moth species composition, richness, alpha diversity and dominance structure. Similarity measures showed that heterogeneity between natural forest and re-vegetated areas was not consistently greater than that within these habitat types, indicating considerable species exchange between sites. Indicator species, defined as those that occurred solely or predominantly in one of the habitat types, comprised approximately 2% of the total assemblage. The ratio of numbers of Pyralidae to the combined total of Geometridae and Noctuidae (the three most prominent families) proved to be a useful index of progression towards a community typical of natural forest. The Misima data will be compared to data from surveys in lowland forest in Madang Province (approximately 1,000 km away), to place these local beta-diversity patterns into a larger-scale context.

Blattodea in the genus *Nocticola* from Australian cave & surface habitats.

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A rich fauna of cave-adapted arthropods, including cockroaches in the genus *Nocticola*, (Blattodea: Nocticolidae) was discovered in the early 1980's in limestone caves of the Chillagoe area and lava tubes of the McBride Formation, North Queensland. This was remarkable because prevailing theories for troglobite evolution predicted that they would not occur in tropical areas. Further discoveries of cave species in Cape Range of Western Australia and across tropical Australia have shown that cave adapted *Nocticola* species are widespread in the Austalian tropics. Search of epigean habitats in the rainforest of east Queensland and in suitable environments inland has revealed surface-dwelling relatives of the cave *Nocticola*. However, the original question remains: are these species the result of allopatric, vicariant evolution or could they have evolved by adaptive shifts in parapatric populations? Major dessication of tropical Australia in the Pliestocene has been substituted for glaciation to explain occurrence of the cave species. Morphological data needs to be used with DNA comparisons to resolve the question of adaptive shift versus allopatric evolution.

The Oniscidea (Crustacea, Isopoda) from groundwater calcretes of Australian arid zones

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Groundwater calcrete aquifers of the Australian arid zone are inhabited by a highly diverse stygofauna, including several species of terrestrial isopods (Oniscidea). Twelve species are presently known: *Anicophiloscia pedisetosa* Taiti & Humphreys, 2001, tentatively included in the family Philosciidae, and 11 blind species in the genus *Haloniscus* (seven from the Yilgarn craton in Western Australia, and four from the Ngalia Basin, Northern Territory). The genus *Haloniscus* includes also *H. searlei* Chilton, 1920, an aquatic species widely distributed in the salt lakes of Western Australia, South Australia, Victoria and Tasmania, and *H. anophthalmus* Taiti, Ferrara and Iliffe, 1995 from anchialine waters in a cave of the Isle of Pines, New Caledonia. All stygobitic species of *Haloniscus* are very similar in external morphology and species separation is mainly possible by examination of the male sexual characters (pleopods and pereopods). A molecular analysis of mitochondrial DNA (CO1) of several populations of the stygobitic species of *Haloniscus* and of *H. searlei* showed that the Australian species form a monophyletic group, while the New Caledonian species is distantly related. The systematic position of the genus *Haloniscus* within the Oniscidea is discussed and the time of divergence of the Australian species of the genus tentatively estimated.

Interaction effects of patch size, edge & stratum on arthropod community on a Japanese oak, *Quercus serrata*, in a fragmented temperate forest

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Arthropods were sampled by branch clipping from two foliage strata (canopy and understory) of Japanese oak trees (*Quercus serrata*) at two forest zones (edge and interior) in two fragments (small and large) of a *Quercus*-dominated secondary forest in Central Japan. In this paper, we focused on interaction effects of fragment size, edge and stratum on four arthropod guilds (chewers, miners, predators and others). The specific aims were to examine 1) whether the impact of edge on guild abundance differs between two foliage strata, exploring a two-factor interaction effect of edge and stratum, and if so, 2) whether fragment size affects the two-factor interaction effect, exploring a three-factor interaction effect of fragment size, edge and stratum. For miners, we distinguished between two groups of weevils (Coleoptera, Curculinidae) and other insects and analysed them separately. Among arthropod guilds and groups, significant two-factor interaction effects on abundance were found in both groups of miners, resulting in less impacts of edge on canopy-dwelling miners than on understory-dwelling miners. A significant three-factor interaction effect on abundance was detected in miners of insects other than weevils, showing the significant less impact of edge on canopy-dwelling miners than understory-dwelling miners only in the large fragment. These results emphasize the importance 1) for the effect of stratum to be taken into account in studies on forest fragmentation and 2) for the edge effect to be evaluated from the viewpoint of interactions with effects of stratum and/or fragment area.

Gall-flies, nematodes & the Myrtaceae: phylogenetic congruence in a tritrophic system

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Flies of the Australasian family Fergusoninidae form galls in a mutualistic association with *Fergusobia* nematodes (Tylenchida: Sphaerulariidae) on various myrtaceous hosts, namely *Angophora*, *Corymbia*, *Eucalyptus, Melaleuca*, and *Syzygium*. Galls are initiated by the feeding of juvenile nematodes deposited by the female fly into meristematic plant tissue during oviposition. Fly eggs hatch and larvae form cavities within the gall into which the nematodes migrate and cohabit with the fly larva. Female nematodes then invade female fly larvae and feed within the haemolymph. Nematode eggs deposited in the fly haemolymph hatch, juvenile nematodes migrate to the fly ovaries, and are inserted into new plant tissue once the female fly emerges from the gall and disperses. It is postulated that speciation in the Fergusoninidae has been driven by radiation of the Myrtaceae and possible incidents of host switching to closely related taxa within it, and that cospeciation between flies and nematodes has been driven by aspects of the fly/nematode life cycle that includes a maternal lineage of fly parasitism, and successive generations of parthenogenetic reproduction and sibling mating in the nematode. In this unique tritrophic system, phylogenetic congruence between flies and nematodes and their myrtaceaeous hosts is investigated.

A review of European temperate forest arthropod canopy studies, using an example from Norway to postulate similarities in host associations & diversity

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We review canopy arthropod studies from four European countries; Denmark, England, Poland and Norway, using available data from published studies in the countries comparatively to assess degrees of host associations and species diversity. A recent study from Norway forms the baseline in the review. Despite geographical differences, differences in sampling methodology and species composition, some similarities were found with respect to host association, faunal structure (e.g. guild composition) and diversity on higher taxonomic levels. We also draw comparisons with studies from North American temperate forests as well as relatively comparable works from the tropics.

Insect diversity in the Great Himalayan National Park Conservation Area, Western Himalaya

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Study on insect diversity along altitudinal gradient was undertaken in one of the globally significant protected sites i.e. Great Himalayan National Park Conservation Area (GHNPCA), Himachal Pradesh, Western Himalaya. The GHNPCA due to its strategic location, and large altitudinal variation (>1,500 to 6,000m) provide a diversity of habitats and associated flora and fauna. Thirty two surveys of average 10 days each covering three different sub-watersheds and 18 intensive study sites in three different seasons were carried out. Only six insect orders *viz.* Coleoptera, Hymenoptera, Diptera, Hemiptera, Odonata, and Lepidoptera were selected for the present study. Assessed insect diversity represented 37 families, 108 genera and 125 species belonging to six orders. The order Lepidoptera represented higher diversity in terms of 55 genera and 61 species followed by order Coleoptera which had 41 genera and 47 species. Out of 37 families, seven (7) families were found in wide altitudinal range (1,500 to >3,500m) while seven (7) families had a narrow distribution (1,500 - 2,000m). Attempt was also made to establish ecological relationships between flowering plants of sub-alpine and alpine areas and corresponding insect visitors. Insect distribution was documented in four broad altitudinal range categories corresponding to major vegetation types *viz.* sub-tropical broad leaved forests (1,500 - 2,000m); temperate mixed conifers (>2,000 - 2,500m); temperate conifers (>2,500 - 3,000m) and alpine pasture (>3,000m).

Rubecula **(Braconidae), introduced biological control agents of** *Pieris rapae*

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In New England (northeastern USA), *Pieris rapae* is an invasive butterfly (ca 1860), against which two braconid parasitoids were introduced: *Cotesia glomerata* (1884) and *Cotesia rubecula* (1988). Neither introduction was preceded by formal host range estimation. Two butterfly species are at potential risk in the region: *Pieris virginiensis* and *Pieris napi*. Recent studies have shown that both are physiologically acceptable hosts for both parasitoids. Field studies showed that the univoltine woodland nature of *P. virginiensis* protects it from attack by either parasitoid. In contrast, a range reduction of *P. napi* appeared to occur shortly after the *P. rapae* invasion. Currently, critical host plants are abundant in both areas retaining *P. napi* (northern New England) and areas now lacking the species (southern New England), and host plants do not explain this range change. Parasitism by *C. glomerata* is high (66-100%) in the summer generation of *P. napi* in both areas retaining and areas lacking the butterfly. The critical difference appears to be a higher rate of commitment to first generation pupal diapause by caterpillars in wooded habitats in northern New England and absence of these parasitoids in wooded areas has protected northern *P. napi* populations, which has avoided parasitism by effectively becoming univoltine in a parasitoid-free habitat.

Effect on native *Pieris* butterflies in New England, USA, by *Cotesia glomerata* and *C. rubecula* (Braconidae), introduced biological control agents of *Pieris rapae*

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In New England (northeastern USA), *Pieris rapae* is an invasive butterfly (ca 1860), against which two braconid parasitoids were introduced: *Cotesia glomerata* (1884) and *Cotesia rubecula* (1988). Neither introduction was preceded by formal host range estimation. Two butterfly species are at potential risk in the region: *Pieris virginiensis* and *Pieris napi*. Recent studies have shown that both are physiologically acceptable hosts for both parasitoids. Field studies showed that the univoltine woodland nature of *P. virginiensis* protects it from attack by either parasitoid. In contrast, a range reduction of *P. napi* appeared to occur shortly after the *P. rapae* invasion. Currently, critical host plants are abundant in both areas retaining *P. napi* (northern New England) and areas now lacking the species (southern New England), and host plants do not explain this range change. Parasitism by *C. glomerata* is high (66-100%) in the summer generation of *P. napi* in both areas retaining and areas lacking the butterfly. The critical difference appears to be a higher rate of commitment to first generation pupal diapause by caterpillars in wooded habitats in northern New England and absence of these parasitoids in wooded areas has protected northern *P. napi* populations, which has avoided parasitism by effectively becoming univoltine in a parasitoid-free habitat.

Life-history traits of invasive phytophagous forest insects

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Exotic phytophagous insects are transforming forest ecosystems worldwide. So far, 36 North American phytophagous insect species have established populations in Europe. 50% of these species belong to the order Homoptera; 17% to Lepidoptera; 14% to Hymenoptera and 11% to Coleoptera. Many of these immigrating species have haplo-diploid or parthenogenetic breeding systems. 75% of the 400 immigrant tree- and shrub-dwelling insects in North America originate from Europe. The same taxonomic order also dominates those European species that have invaded to North America. However, the proportion of European coleopterans that have invaded to North America is two times higher than that from North America to Europe. No North American hemipteran species has been able to colonize the European continent, but 13 European hemipteran species have successfully invaded to North America. The majority (56%) of North American phytophagous insects in Europe live on deciduous trees. 20% of those host plants are of North American origin. 44% of the invaded species live on conifers and 63% of the coniferous host plants are of North American origin. Of the European emigrants in North America, only a few have been able to colonize native North American plant genera. This is especially true among homopterans feeding on conifers.

The composition of mangrove ant communities as indicator for human impacts of a wildlife refuge in Taiwan

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The Wildlife Refuge is one of five categories of protected area system in Taiwan. Since central and local governments share the responsible authorities of Wildlife Refuges, the effectiveness of the protection and maintenance of biodiversity is limited. The Wildlife Refuge which the study was carried out, located in southwestern coast of Taiwan and famous for its diverse mangrove plants and migratory birds, is confronted by human interferences, such as traditional fishery uses of local people and intensive domestic tourism activities. To understand the impacts of human activities on the territorial areas of the mangrove system, the survey was conducted during October 2002 to September 2003 by using food baits and pitfall traps to collect ants monthly. Based on sampling in two different management areas which were also divided into four levels of human disturbances, the community structure and species richness of ants were determined. There were a total of 58 ant species collected in the Wildlife Refuge, and there were differences of ant abundance and species richness between study sites and between months. Species richness and ant abundance were highest in October and lowest in January. The more the management involved and the less human disturbance level the site, the more was the species numbers and more species supported.

Pattern and process in subterranean speciation: comparative analysis of Australian and Hawaiian cave planthopper radiations (Hemiptera: Fulgoromorpha: Cixiidae)

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Ongoing studies have revealed numerous cases of parallel evolution of cavernicolous planthoppers all over the world in tropical and subtropical caves. Given that concepts of genetic change and speciation dynamics are subject to review by natural model systems, the comparative study of these independent evolutionary lineages and radiations can provide critical information. We survey species complexes from Australian and Hawai'ian caves exhibiting different degrees of troglomorphy. In Australia (Qld), *Solonaima* and *Undarana* species have colonized old karst caves as well as young lava tubes. On the Hawai'ian Islands, the cave-dwelling *Oliarus* species represent at least seven independent cave colonizations on three islands of different age, including the *Oliarus polyphemus* species complex from various, partly still active volcanic systems on Hawai'i Island . The diversity of external factors (e.g., ressource availability and stability, migration possibilities, macro- and microclimatic changes, predators) as well as internal factors (e.g., genetic variability, population structure and density) allows us to incorporate these into concepts of evolutionary change. On this basis we explore verbal models of cave adaptation (Relict hypothesis sensu BARR, Adaptive shift hypothesis sensu HOWARTH) and analyze the influence of sexual selection and founder effects sensu MAYR and CARSON.

Paradise lost? A long-term term perspective on species endemicity, rarity & extirpation using the fossil Coleoptera record

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This paper examines the dimension provided by the study of fossil insects (mostly beetles) preserved in waterlogged deposits in bogs and archaeological sites to study long-term changes to our insect fauna. It examines the contribution of the fossil insect record to our understanding of species evolution and what it can tell us about endemic species, rarity and extirpation. The paper draws upon examples from northern Europe and in particular upon the author's research from lowland raised bogs in the Humberhead Levels, eastern England, where the particular faunal characteristics, including its endemics, of these lowland raised mires were already in place 4000 years ago. Additionally, research indicates that over 38 species of Coleoptera (mostly saproxylics) have become extirpated from the British Isles within the last few thousand years, probably due to a combination of forest clearance and human activities, ainage of wetlands, edaphic and competition factors affecting distribution of host trees, climate change, forest fire decline and a decline in grazing. The fossil insect work provides a long-term perspective on the effects of such factors upon the beetle fauna and most of all, highlights the important part played by time and history in shaping the nature of our fauna today.

What can forest managers learn from research on subfossil insects?

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This paper outlines the usefulness of using fossil insects, particularly Coleoptera, preserved in waterlogged deposits to understand the changing nature of forest ecosystems and their associated insects over the last 10,000 years. Research in Europe has highlighted the complex nature of early woodland ecosystems. In particular, the role of dead wood as well as grazing animals will be discussed, suggesting that the north European primary forest has similarities to pasture woodlands, rather than the forester manager's perception of closed canopy. Later on, human activity had a major impact on these ecosystems, with the concurrent expansion of plants and animals associated with cleared landscapes and pasture and resulting in the local extinction of a sizeable proportion of taxa from the forests of northern Europe. The decline in these species has been seen as resulting from habitat loss due to human forest clearance, which intensified from about 2500 years ago onwards, coupled with subtle climate change effects. These extirpations will be discussed, with particular reference to the management of forest ecosystems for the benefit of their invertebrate species.

The effects of the exotic weed Bitou Bush (*Chrysanthemoides monilifera* ssp. *rotundata* L.) on terrestrial arthropod communities in a littoral habitat

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This study investigates the impacts upon selected arthropod assemblages of habitat invasion by bitou bush (*Chrysanthemoides monilifera* ssp. *rotundata*). Sixteen sites were placed at four geographic localities on the coast of New South Wales. The sampling design had two spatial scales (between and within localities) and eight repeat temporal measures. Arthropods were collected from a variety of microhabitats using several sampling techniques, and specimens of ants, beetles, true bugs and spiders were identified to species level. Differences in α diversity and population abundance structures between the taxonomic assemblages are described, along with comparisons of data contrasts between bitou-free ("control") and bitou-invaded ("impact") habitats and between geographic localities. A subsidiary impact associated with herbicide application for control of the weed is also examined. Univariate diversity descriptors detected some differences between control and impact treatments within a locality, however they are not as apparent as between geographic localities. Constrained ordination analyses detected evidence for associations between arthropod assemblages and particular vegetation types. They also demonstrated arthropod community composition change that appeared to be related to herbicide application for some taxonomic groups.

Rot type, log size and forest type; characteristics that influence saproxylic beetle assemblages

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In Tasmania, as seen in Northern Europe, saproxylic communities are at risk of being negatively affected by intensified forest management that results in the removal of oldgrowth elements, such as late successional forests and large diameter logs. As there is a shorter history of forest management in Tasmania, there is still time to reduce any ecological impacts through understanding characteristics that influence beetle assemblages. We intensively sampled saproxylic beetles and rot types within decomposing *Eucalyptus obliqua* logs in Tasmania's wet eucalypt forests. We compared the beetle assemblages between large (>100 cm) and small (30-60 cm) diameter logs, in multi-aged unlogged and logging regenerated forests. Logged forests support a diverse but significantly different beetle assemblage to multi-aged unlogged forest, possibly representative of early successional forest. Large logs host a distinct suite of beetles not found within small logs. As indicated by their rot types large and small logs appear to undergo different decomposition processes, explaining, in part, the distinct faunas associated with different log sizes. Results are discussed in relation to addressing conservation issues within a production forestry landscape.

Can tree planting be a threatening process for invertebrates?

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Since European settlement in Australia, large scale tree clearing for agriculture and urban development has resulted in significant losses in the biodiversity of native plants and animals, although some native species have benefited from this. Major initiatives to replant lost trees have presumed that tree planting will enhance native biodiversity. This approach is necessary and appropriate, and in the main, will enhance invertebrate biodiversity primarily through provision of food and habitat. There could, however, be environmental harmful effects with biosecurity implications. Some of these, such as the introduction of exotic species of invertebrates, are readily apparent, but some are more subtle. Tree planting may change the host plant relationships of endemic invertebrates by altering the composition of insect assemblages on these plants, and this has potential evolutionary implications. The insect assemblages on *Eucalyptus* and *Acacia* have a unique coevolutionary history in Australia, and this could be severely disrupted or altered by inappropriate selection of plant species. Even the well-meaning intension of planting native flora, requires care in the choice of plant chosen in revegetation projects.

Genetic structure and gene flow in island populations of pollinating fig wasps

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Pollinating fig wasps (*Hymenoptera, Agaonidae*) are associated with fig trees (*Ficus, Moraceae*), a keystone plant species for the functioning of many tropical ecosystems. The Krakatau islands in Indonesia are an ideal natural experiment site to study colonization and gene flow processes in disturbed and fragmented habitat, as they were completely sterilized by volcanic eruption in 1883. Since then, vegetation has been returning to the islands in a successional manner, with several *Ficus* species being pioneer species. However, pollinator limitation has been observed in some populations of dioecious *Ficus* on the Krakatau islands. We used recently developed microsatellite markers for pollinating fig wasp *Liporrhopalum tentacularis* (pollinator of dioecious *F. montana*) to estimate genetic differentiation between islands and surrounded Indonesian mainland of Java and Sumatra. Our results indicate that genetic diversity in the investigated pollinating fig wasp on the island is similar to that on the mainland and gene flow is not restricted between island and coastal sites of the mainland. However, gene flow between Krakatau islands and distant Java inland sites was found to be more restricted. Consequently, we discussed wasp dispersal and the implications of our findings on the maintenance of the fig – fig wasp mutualism.

Biodiversity of tropical rainforest bark beetles & timber borers: results from an intensive inventory

L Kirkendall

Paper not available at time of production

&The phylogenetic position of Sericini examined by morphology based cladistics (Coleoptera: Scarabaeidae).

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In contrast to the coprophagous Scarabaeidae, the phylogeny of the of phytophagous Scarabaeidae is far from being resolved. I produced a cladistic analysis to investigate the basic phylogenetic relationships of Sericine chafers and the systematic position within the family Scarabaeidae employing 107 morphological characters of 49 extant scarabaeid genera. These taxa represent most of the 'traditional' subfamilies of coprophagous (only two subfamilies included) and phytophagous Scarabaeidae. The cladogram obtained from the parsimony analysis confirms a monophyly of phytophagous Scarabaeidae (including e.g. Orphninae, Cetoniinae, Dynastinae, Euchirinae, Rutelinae, Melolonthinae). While the respective monophyly of Cetoniinae and Rutelinae is well supported, the Melolonthinae result paraphyletic, since both former groups are nested within melolonthine lineages. The Ablaberini were shown to be the sister group of the Sericini at the top of a clade, with mainly pectinate branching including tribes of southern continents (e.g. Diphucephalini, Scitalini, Heteronycini). Preliminary results obtained from the phylogeny and knowledge of recent distribution of the clades favour one hypothesis for the evolution of the Sericini: the evolution of main lineages have taken place on African continent supposingly after the splitting of the Gondwanian continent and the ancestor of Neotropical Sericini dispersed during Tertiary over the atlantic ocean.

Polypedilum (Diptera: Chironomidae) from Egypt with description of a new species

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A survey of chironomid midges (Diptera: Chironomidae) conducted from September 1997 to October 1999, covering all major geographical zones in Egypt, resulted in the capture of 5 species of the genus *Polypedilum* Kieffer. Three of these species are new records, while one new species, *Polypedilum (Uresipedilum) marsafae*, is described from male and female imagines. The Egyptian list of *Polypedilum* Kieffer is updated to include 11 species. A key to the male imagines is presented. New distributional data for the species encountered are also given.

Abdominal morphology of the Protura (Hexapoda) and phylogenetic implications

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Cladistic analyses of arthropod phylogeny presently support the view that insects are closer related to crustaceans than to myriapods. Available data, however, neither permit to specify the insect sister group, nor do they significantly support the monophyly of the Hexapoda (= Insecta *s. lat.*). Instead, the relationships of the basal subgroups remain unclear that are traditionally united as "Entognatha" (Collembola, Protura, Diplura). In order to contribute to a clarification, we started a comparative study on the morphology of the abdomen in primarily wingless insects with emphasis on the skeleto-muscular as well as nervous system. Main focus is targeted at the Protura as these seem to be less derived relative to all other insects in maintaining anamorphic development and articulated appendages at the first three abdominal segments. Details on the morphology of the proturan pregenital abdomen are presented based on a combined approach by transmission electron microscopy and confocal laser scanning techniques. This approach permits to establish hypotheses on the primary homology of the abdominal muscles by the criteria of topography and innervation. Preliminary insights derived from the results obtained are discussed in the light of the assumption favoured by some molecular data that the Hexapoda form a paraphyletic assemblage.

The evolutionary history of insect storage hexamerins and phylogeny of the insects

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Insect hexamerins are large hemolymph proteins (~500 kDa) that are related to arthropod hemocyanins, but have lost the ability to bind oxygen. They are composed of six similar or identical subunits. Hexamerins may accumulate to extraordinarily high concentrations, usually in larval or nymphal stages. Some hexamerins are rich in methionine ("methionine-rich storage proteins") or aromatic amino acids ("arylphorins"). Most hexamerins are considered mainly as storage proteins that provide amino acids and energy for non-feeding periods, e.g. pupal stages. However, in recent years, other specific functions like cuticle formation, transport of hormones or humoral immune defense have been proposed. Hexamerins emerged from hemocyanins early in insect evolution. The phylogeny of the hexamerins has been reconstructed based on a multiple sequence alignment, employing distance methods, maximum parsimony and Bayesian inference. The data show that the hexamerins evolved in line with the phylogeny of the insect orders. Within the orders, there is a notable diversification of these proteins into various subtypes, which probably reflects specific functions.

On the non-homology of several phallic "spiculae" of Plant Bugs (Heteroptera, Miridae).

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Male genital structure has been used in the taxonomy of the Miridae for nearly 100 years. The importance of male genitalia to the diagnoses of plant bug taxa is obvious. However, the terminology used to describe these structures remains plethoric and ambiguous. This situation is the result of a combination of structural complexity and absence of agreement by specialists on homology assumptions for these diverse structures. This terminological ambiguity is well illustrated by the term phallic "spiculum." Currently reference to this term in the taxonomic literature applies to three different non-homologous structures. These structures are illustrated and briefly described.

A new species of *Prosopistoma* (Ephemeroptera: Prosopistomatidae) from Myanmar

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A new species, *Prosopistoma konanwina* (Ephemeroptera: Prosopistoma) is described and illustrated, based on larval material from Mon state, Myanmar

Morphological and mitochondrial DNA marker analysis of burrowing mayflies, *Ephemera (Ephemeroptera: Ephemeridae) in Korea*

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Morphological characters of the burrowing mayflies, *Ephemera* spp. (Ephemeroptera: Ephemeridae) in Korea were studied using scanning electron microscopy. Random amplified polymorphic DNA polymerase chain reaction markers proved complementary to the morphological identification of the burrowing mayflies. From each species of the burrowing mayflies, a region of the mitochondrial DNA COI gene was amplified, cloned and the sequence determined. Parsimony and distance analyses were performed and the results were similar to those based on morphology. The combination of morphological and molecular traits is useful in understanding the diversity and evolution of these burrowing mayflies.

Systematic revision of the Cossulinae (Lepidoptera: Cossidae) of Costa Rica

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The Cossidae is an economically important family consisting of six subfamilies and more than 700 species globally. Their larvae are often of serious concern to forestry and horticulture in that they are known to bore in the branches and trunks of a wide range of trees and shrubs. In the course of my examination of the extensive collections of the Smithsonian Institution and the Instituto Nacional de Biodiversidad of Costa Rica, I have found the Costa Rican Cossulinae to consist of 17 species, including 8 new to science. The phylogenetic relationships of all cossuline genera, including 4 new genera, will be analyzed using the computer programs PAUP, WINCLADA and NONA. Data for the study has been derived from extensive microscopic slide preparations and examinations of both external (body, leg, and wing morphology) and internal (primarily genitalic morphology) anatomical structures. Diagnostic characters of all taxa have been photographed using digital equipment involving automontage, supplemented with camera lucida drink drawings for additional detail where necessary. Diagnostic descriptions and identification keys accompanied by detailed illustrations of every species are being generated, and the recorded distribution of each species will be clearly displayed using topographic maps of Costa Rica.

Phylogenetic relationships among species of the genus *Stelis* in North America (Hymenoptera: Megachilidae)

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The genus *Stelis* comprises a colorful group of parasitic Anthidiine bees that are seldom encountered or collected at floral resources. Adults spend most of their time searching for hosts or waiting near host nests until they are available for parasitism. Like their parasites, all hosts are in the family Megachilidae. Herein, we describe 65 North American species in this Holarctic genus, including twenty new species, and analyze the group with respect to morphological characters, host, and host nest characteristics by parsimony analysis and successive approximation character weighting.

The genus *Acrosternum* Fieber, subgenus *Chinavia* Orian (Hemiptera, Pentatomidae, Pentatominae) in Brazil: systematics, distribution, host plants, immatures and bionomics.

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The genus *Acrosternum* includes a significant number of the so-called "green stink bugs" which are widespread in the world, except in the Australian region. Many species of *Acrosternum* are associated with agroecosystems. Two sub-genera are recognized: *Acrosternum*, with 15 spp., and *Chinavia* with 77 spp. More than sixty species, belonging to *Chinavia*, are found exclusively in the neotropics, testifying the great species diversity in this region. The monophyly of the group has been discussed, and this subject is under investigation at our laboratory, including the revision and biogeography of *Acrosternum*. About 30 species are known from Brazil, near fifty percent described in the last two decades; five of them were recognized as knew by the authors. At least ten species are associated with important agricultural crops as cotton, green-beans, soybeans and tomatoes, but information on biology and natural history of those species are relatively unknown. In the present study the systematics, geographical range, host plants, morphology of immature stages and bionomics of the species recorded to Brazil are discussed, based mostly on unpublished data, as in literature.

Two new species of Heliozelidae (Lepidoptera) associated with myrtaceaous plants from southern China

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The family Heliozelidae includes very small-sized moths with forewing length of 3-9mm, comprising 12 genera and about 100 species in the world. It has a worldwide distribution mainly in the Nearctic and Australian regions, but it has not been known from South East Asia. In March and December 2003, participating in a research project "Network construction for the establishment of insect inventory in Tropic Asia (TAIIV: organized by O. Yata of Kyushu Univ.)", we collected larvae of 2 unknown heliozelid species mining on plants of Myrtaceae in southern China (Guangzhou and Hainan Is.). Larvae of these species have different types of mines. One (Heliozelidae sp. 1) mines into the miib of *Syzygium jambos*, and the other (Heliozelidae sp. 2) into the blade of *Rhodomyrtus tomentosa*. Examination on morphological characters of emerged adults showed that Heliozelidae sp. 1 was assignable to *Heliozela* but Heliozelidae sp. 2 could not be placed in any described genera. As regards the host plant family, Nielsen (1985) suggested presence of a heliozelid species of unnamed genus associated with myrtaceaous shrubs and trees in South America, but the species has not been described. Discovery of heliozelid species associated with myrtaceaous plants in southern China may indicate their Gondowanian origin.

Structure and function of bristle-like sensilla on the maxillary palp of *Priacma serrata* (Coleoptera: Archostemata)

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Priacma serrata LeConte, 1861 is a member of Cupedidae, the most ancient taxon of Coleoptera. Scanning electron micrographs show that there is a clearly defined sensilla area on the apical segment of the maxillary palp of *P. serrata* and all other archostematan beetles. In *P. serrata* this area contains 10 to 17 erect bristle-like sensilla. In other Coleoptera, Adephaga as well as Polyphaga, there usually is a field of characteristic digitiform sensilla in the same area of the apical palpomere. Each of these sensilla is counter-sunk into an elongate depression of the cuticula of the palp. The function of these sensilla is still unknown. One aim of our investigation of the ultra-structure of the bristle-like sensilla of *P. serrata* was to answer the question, whether these and the digitiform sensilla of other Coleoptera are homologous structures. The second aim was to identify the function of these sensilla. So far the study shows that the bristle-like sensilla have numerous, deep, longitudinal grooves and no apical porus. Each sensillum is moveably inserted into the cuticula and innervated with a single sensory cell. This combination of characters is indicative of a chemo-sensitive olfactory receptor.

The wing base of Ephemeroptera and the ground-pattern of insect flight

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The ability to fly is the decisive factor for the evolutionary success of winged insects (Pterygota). Despite this, very little is known about the ground-pattern and evolution of the functionally very important wing base. This research project endeavours to contribute to the knowledge of the evolution of the flight system of the insects. The skeletal elements of the wing base, as well as the associated muscles are examined. For this purpose, in addition to classical methods, micro-x-ray-computer-tomography is used. This method enables an investigation into the muscular and skeletal systems and a straightforward reconstruction of virtual 3D models without any artefacts. The results so far include new findings regarding the flight musculature of the Ephemeroptera and the homologisation of individual sclerites of the wing bases of Ephemeroptera and Neoptera. The final goal of this research project is to establish the ground-pattern of the axillary areas of the wings of the Pterygota. Furthermore, the investigation of the wing apparatus will provide important information for the analysis of the phylogenetic relations between individual taxa of the Ephemeroptera.

Molecular phylogeny among the subfamilies of Ichneumonidae (Hymenoptera) inferred from the COI gene sequences

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A phylogenetic study among four subfamilies of the Malaysian Ichneumonids (Hymenoptera) were examined using the complete mitochondrial cytochrome oxidase I (COI) gene sequence data. A total of 1688 base pairs of the COI gene were sequenced from 23 individuals comprising the subfamilies of Anomaloninae, Campopleginae, Cryptinae and Metopiinae. Members of the subfamily Braconidae were selected as an outgroup. Phylogenetic analysis was conducted using both the Maximum Parsimony and Neighbor-Joining methods. The Maximum Parsimony analysis showed the Ichneumonidae group to be monophyletic with two major clades; I) a clade containing members of Campopleginae and Anomaloninae and II) a clade containing members of Cryptinae and Metopiinae. The close relationship of Campopleginae and Anomaloninae is supported by previous morphological classifications which classified them into the Ophionoid group. However, the relationships of Cryptinae and Metopiinae is not supported by the previous studies as they belong to a different subfamily group. A broader sampling effort throughout Malaysia should be made to provide a clearer picture of the genetic differentiation within this family.

A world revision of the genera of the family Scelionidae (Hymenoptera: Platygastroidea).

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The family Scelionidae encompasses 165 genera and 3308 species, and is found worldwide. All species are egg parasitoids of other arthropods, attacking hosts in the Araneae, Odonata, Orthoptera, Mantodea, Embiidina, Hemiptera, Coleoptera, Neuroptera, Lepidoptera, and Diptera. A revision of the genera of the world is underway, redefining known genera based on extensive new collections and the examination of type species; describing new genera; and documenting the results via an on-line accessible database. Recent results: The genus Thoron Haliday was known from only a single Holarctic species, parasitic in the eggs of Nepidae (Hemiptera). The limits of the genus have been redefined and the name Neothoron Masner is considered a junior synonym. The genus Plaumannion is described as new, with two species in South America. This taxon lacks one of the fundamental characters of the family and may merit recognition at a higher taxonomic levelResolution of the question is part of the ongoing phylogenetic analysis of the group. The species and monophyly of the putatively basal genera Nixonia, Sceliomorpha and Archaeoscelio are being revised.

A new tribe and two new genera of Eulophidae (Hymenoptera) from West Australia

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A new tribe of gall associated wasps is described from West Australia to contain two new genera. One genus is associated with plant galls on *Melaleuca* (Myrtaceae), and may be phytophagous; the biology of the second genus is unkown. The tribe is considered to be most closely related to a group of tribes with includes Ophelimini, Anselmellini (both phytophagous) and Platytetracampini (parasitoids of whiteflies). Morphological characters to support the monophyly of this tribe are provided, and include: antenna placed very low on the face, submarginal vein with only a single dorsal seta, and antenna with 4-5 small segments between the pedicel and club which are not clearly differentiated into anelli and funicle. Both new genera display morphological adaptations associated with extremely long ovipositors.

Phylogenetic reconstruction of black flies (Diptera: Simuliidae) from Thailand, using ITS2 rDNA

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The sequence of internal transcribed spacer (ITS2) of ribosomal DNA (rDNA) was used to estimate the phylogenetic relationships of 40 black fly species belonging to at least 4 subgenera of the genus Simulium: Gomphostilbia (12 species), Nevermannia (5 species), Montisimulium (1 species) Simulium sensu stricto (21 species), and an unknown subgenus (Simulium baimaii). Maximum parsimony (PAUP version 4.0b10) analysis revealed two major monophyletic groups. The first monophyletic group consists of Gomphostilbia and Nevermannia + Montisimulium. The second monophyletic group consists of the subgenus Simulium and one species of an unknown subgenus. (Simulium baimaii). The molecular phylogeny in the present study generally agrees with existing morphology-based phylogenies.

Phylogenetic relationships of *Megoura* species (Hemiptera: Aphididae) in the world

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The genus *Megoura* Buckton, 1876 consists of 7 species in the world, which are related with Leguminosae. A total of 42 morphological characters are selected and two DNA markers, mt COII (mitochondrial cytochrome oxidase subunit II) and EF1α (elongation factor 1 alpha) are sequenced for the phylogenetic analysis. All of the 7 species are used in the morphological analyses, but 2 species (*M. litoralis* F. P. Müller, 1952 and *M. dooasis* (A.K. Ghosh & D.M. Raychaudhuri, 1969)) are not available for the molecular phylogenetic analyses. When analysed with maximum parsimony and maximum likelihood methods and rooted with *Acyrthosiphon* species on Leguminosae species, both independent and combined phylogenetic trees are coincident with and support the previous discussion based on morphology (Lee et al., 2002). This results also elucidate the taxonomic status of the three similar species (*M. viciae* Buckton, 1876, *M. crassicauda* Mordvilko, 1919, and *M. nigra* Lee, 2002) on the herbaceous Leguminosae, *Vicia* Tourn. Ex Linn. and *Lathyrus* Linn. Both morphological and molecular results support that *M. crassicauda* in the Eastern Palaearctic, the former subspecies of *M. viciae* Buckton, 1876, is very close, almost identical to *M. viciae* in the Western Palaearctic, while *M. nigra* Lee, 2002 in Korea is distinctively separated from the other two species. The 7 world *Megoura* species are divided into two clades, one group consisting of *M. crassicauda*, *M. viciae*, *M. nigra*, and *M. litoralis*, on the herbaceous legume plants and the other group of *M. dooasis*, *M. lespedeza* (Essig & Kuwana, 1918), and *M. brevipilosa* Miyazaki, 1971, on the woody legume plants (*Hedysarum* Linn., *Indigofera* Linn., *Cajanus* DC., *Desmodium* Desv., *Lespedeza* Michx.). This implies that the speciations of *Megoura* species have occurred in the close relationship with the host plant radiations, although the evolutionary relationships of the host plant genera in Leguminosae have not been solved clearly yet.

A taxonomic study of Chinese phorid flies(Diptera: Phoridae)

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China, meeting both the Palearctic and the Oriental region, is rich in phorid fly resources. At present study 87 species of phorid flies in 22 genera are recorded, of which 22 species are new to science , 7 genera and 4 species are reported for the first time in China. The following genera are covered: *Hypocera* Lioy, *Borophaga* Enderlein, *Latiborophaga* Brown, *Godavaria* Brown, *Chaetogodavaria* Liu, *Ctenopleuriphora* Liu, *Stichillus* Enderlin, *Phora* Latreille, ,*Spiniphora* Malloch, *Psyllomyia* Loew, *Diplonevra* Lioy, *Dohrniphora* Dahl, *Conicera* Meigen, *Chouomyia* Liu, *Triphleba* Rondani, *Rhopica* Schmitz, *Metopina* Macquart, *Trophithauma* Schmitz, *Puliciphora* Dahl, *Gymnophora* Macquart, *Woodiphora* Schmitz, *Phalacrotophora* Enderlein. Keys to genera and species are presented to facilitate identification. All the genera and species are described or rediscribed.

The Neotropical genus *Rotundicerus* Maldonado-Capriles, 1977 (Hemiptera: Cicadellidae: Idiocerinae)

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The genus *Rotundicerus* was described by Maldonado-Capriles (1977, Proc. Entomol. Soc. Wash. 79(3): 317-325), with its type species *R. luteus* Maldonado-Capriles from Guyana. This genus is characterized by the vertex finely shagreen and its anterior and posterior margins strongly curved and parallel, forewings with the inner anteapical vein without a cross vein, aedeagus without socle and caudal side on apical half with scaly appearance, pygopher triangular in lateral profile, and anterior dorsum mostly yellow. Dietrich (in Dietrich & McKamey, 1990, Proc. Entomol. Soc. Wash. 92(2): 214-223) described two more species, *R. minutus* Dietrich and *R. rubripictus* Dietrich, from Guyana. In this paper, four more species of this genus are added, two from Peru (Jenaro Herrera, Loreto) and two from Brazil (Jacareacanga and Santo Antonio de Taua, Para). Based in the study of this additional material, the two species described by Dietrich are excluded from *Rotundicerus*, representing species of two undescribed genera.

Diperocarp seed predation by weevils: succession and specificity (Coleoptera: Curculionidae)

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Dipterocarp trees dominate much of South-East Asian forests. There are more than 400 species of the family, and species grow in mixed stands. The fruit is attacked by a number of beetles and moths, and mortality of up to 100% has been recorded. Although traditionally seed predators have been grouped into 'pre-dispersal' and 'post-dispersal' this categorisation does not capture the successional aspect of weevil attack on dipterocarp seeds, where time of oviposition is allied to physical condition of the developing fruit and whether the fruit is on the tree or not on emergence of the adult beetle. *Alcidodes* weevils show specificity at the level of host genus or species group, although quantitative studies show a host preference within the range, while Nanophyinae show specificity at the generic or species level. Host tree individuals can have several congeneric beetles attacking the fruit.

LOWD: Leafhoppers of the World Database

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An electronic, relational species database of world leafhoppers (Cicadellidae) is nearing completion using the software program *Biota* 2 (Colwell 2004). Presently there are 18,972 valid species and subspecies among 2,609 valid genera and subgenera. The database includes the current taxonomy, synonymy, and taxonomic history since 1955 for 21,769 available species names, including type depositories for 13,447 and country-level distribution for 20,302. Data is derived from Metcalf's Catalogue of the Homoptera (10,952 species, coverage 1758-1955), Oman, Knight & Nielson's 1990 generic checklist and bibliography, the Zoological Record, and original descriptions (10,035 species, 1,397 papers). Only 782 databased species names remain to be checked against their original descriptions and perhaps 1,500 species have been described but not yet databased. LOWD is also exceptionally well suited for specimen data and collection management. Within this year, specialists will be invited to import specimen records directly from remote stations to establish an unparalleled directory of specimens available for research. Within five years, the periodically updated database will be posted for free access on the WWW. Receipt of publication lists and reprints of taxonomic papers on Cicadellidae (and other Auchenorrhyncha) will be appreciated most gratefully and will ensure their incorporation into the database.

Liposcelis bostrychophila **Badonell: RAPD fingerprinting pinpoints its geographic origin**

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Liposcelis bostrychophila (Psocoptera: Liposcelididae) is a cosmopolitan pest of stored grain in Australia and internationally. It has become a pest due to its parthenogenetic life history and tolerance to adverse environmental conditions. As there is currently limited information on the invasion biology of *Liposcelis* species, a greater understanding of the general population biology of *Liposcelis* species, including population structure and dynamics, gene flow among populations, geographic origin of source populations and dispersal mechanisms is required. Elucidation of genetic structure in geographic populations is an important aspect of pest studies providing insight into the geographic origin of colonised populations. *L. bostrychophila* is thought to have dispersed globally via commerce. It is suspected that *L. bostrychophila* has been introduced to Australia, but from unknown sources. To define the origin of pest populations, genetic markers can be used to test for levels of genetic similarity. This poster details the use of Randomly Amplified Polymorphic DNA (RAPDs) to elucidate the geographic origin of *L. bostrychophila* populations in Australian grain storage systems.

The higher classification of cicadas: a tentative cladogram

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Opinions on family and subfamily groupings within the Cicadoidea remain varied. A broad consensus has emerged for the recognition of just two families, the Tettigarctidae comprising mostly fossil genera and two extant species, and the Cicadidae which encompasses all other living cicadas as well as fossil taxa. Traditionally the large majority of Cicadidae have been divided into subfamilies, the Cicadinae and Tibicininae. The status of other groupings, particularly those based on *Plautilla* and *Tettigades,* as either subfamilies or families remains in contention. I have tested the validity of all family and subfamily groupings using cladistic parsimony. Support for just two family groupings, the Tettigarctidae and Cicadidae was very strong. The subfamily Cicadinae, those cicadas of which the males possess timbal covers was also supported, as were the Tibicininae. A notable transgression from current thinking was the monophyly of *Platypedia, Tettigades* and *Tibicina.* This group falls as the sister group of all other Cicadidae and is proposed as the new subfamily Tettigadinae. One further subfamily grouping is identified, proposed as the Zammarinae. The monophyly of the large tribe Cicadettini, based upon the partial fusion of fore wing veins M and CuA, is challenged. The Cicadettini are here considered to include some genera lacking fusion of these veins.

Proteininae (Coleoptera: Staphylinidae) in Gondwana: ancient distribution patterns?

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Prior work and ongoing studies indicate that the subfamily Proteininae is a monophyletic, basal lineage within the huge beetle family Staphylinidae, and consists of five evidently monophyletic tribes of two or three genera each. Four tribes are restricted to austral or Gondwanan regions, while the fifth and largest (Proteinini) is most diverse in the northern hemisphere but includes species in tropical regions and a few on the fringes of southern temperate areas. Among the four austral tribes, earlier taxonomic work based on a few dozen known specimens identified a total of 8 genera and 11 species, but systematic review of abundant material from recent and continuing field surveys indicates the presence of at least one new genus and dozens of new species, and significant range extensions for each group including new trans-austral connections. All four austral tribes now show southern connections between three areas (Silphotelini and Austrorhysini, each found in Australia, New Zealand and southern South America) or two areas (Anepiini in Australia and New Zealand, and Nesoneini in New Zealand and New Caledonia). Continuing generic and species-level systematic revisions of the austral tribes attempt to elucidate phylogenetic relationships among all taxa and analyze global and local distribution patterns.

Higher phylogeny of Zygaenidae (Lepidoptera) and evolution of chemical defence

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The higher phylogeny and evolution of chemical defence mechanisms of Zygaenidae are investigated using morphological characters and 4 molecular markers (18S rRNA, 28S rRNA, ND1, 12S rRNA). Both morphological and molecular evidence show that the Phaudinae does not belong to the Zygaenidae because it forms a clade with Lacturidae, which in turn is shown to be the sister-group of the limacodid-family group. Anomoeotinae and Himantopteridae also do not belong to Zygaenidae because the clade comprising these two families is the sister-group of Somabrachyidae. Within the Zygaenidae, the morphological and molecular trees show significant topological incongruence. The morphological phylogeny shows that Procridinae is the most basal zygaenid group and the Zygaeninae is the sister-group of (Callizygaeninae + (*Heteropan* + Chalcosiinae)). However, the molecular phylogenies suggest that the Zygaeninae forms the most basal group, with the remaining subfamilies forming an unresolved polytomy. According to the morphological phylogeny, absence of the cuticular cyanic fluid storage system in the larvae is supposed to be a plesiomorphic state, but the molecular phylogeny indicates that absence of this mechanism may be due to a secondary loss. We also compare the parasitoid faunas associated with the larvae of different zygaenid groups to see if there is significant difference in host use under different chemical defence systems of the host caterpillars.

Life stages and biology of *Filinota brunniceps* (Felder & Rogenhofer) (Lepidoptera: Elachistidae: Depressariinae) from Costa Rica

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For over 100 years, *Filinota brunniceps* (Felder & Rogenhofer) was known only from a single adult specimen from Colombia SA, until it was rediscovered in Mexico and later by K. Nishida in Costa Rica. All life stages for this species, its biology, and some larval and adult behaviors are described here. Larvae feed on leaves of *Rivina humilis* (L.) [Phytolaccaceae]. Eggs are ovoid with a transverse medial ridge slightly beyond each end margin. Eggs are usually laid in grooves of the host plant stem. Larvae live as leaf scraper then rollers. Pupation occurs within rolled leaf away from the host plant. The pupa is deeply excavated laterally between the labrum and the proximal margins of the maxillae and possesses a ridgelike "cocoon cutter" on the dorso-lateral area of the head. Adults are diurnal to crepuscular and exhibit an unusual survival behavior for Microlepidoptera. When at rest, conspicuous adults rolls their wings around their abdomen, exposing an "eye spot" on each side of the posterior end of the forewing while the terminal portion of the antennae extend well beyond the posterior wing margins, giving a "false head" search image to would be predators.

Moving toward Thysanopteran phylogeny: DNA extraction methods that preserve voucher specimens

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Our ultimate goal is to estimate thrips phylogeny using morphological and molecular characteristics. Reliable thrips identification using morphological methods requires use of a compound microscope to view cleared specimens mounted on slides. For molecular phylogenetic research, it is essential that specimens from which DNA is extracted are identified accurately. However, preparation for molecular analysis is usually destructive, and identification based on other thrips from the same collection may be confounded by the presence of more than one species in a sample. To avoid such misidentifications, we developed a technique that preserves the thrips exoskeleton for mounting as a morphological voucher, while extracting strong DNA product for our molecular analysis. We make a small incision along the abdomen, and place the whole thrips into a Qiagen^{TM} DNeasy tissue kit, proteinase K lysis buffer (ATL) for processing. We do not grind up the specimen. The thrips exoskeleton is then retrieved from extraction kit, dehydrated and mounted in Canada balsam on a microscope slide. We also extracted thrips DNA using the methods of Sunnucks and Hales (1996) and D.C. Morris (*pers.com.*), but obtained lower DNA yields than with our method.

Phylogeny of the Hippoboscoidea (Diptera), based on molecular characters.

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The Hippoboscoidea is a superfamily of Diptera that include Glossinidae (tsetse flies), Hippoboscidae (keds and louse flies), Streblidae and Nycteribiidae (bat flies). All members are blood feeders of mammals or birds. While the Glossinidae are free-living, the remaining members of the group are all obligate ectoparasites, spending most or all of their life-cycle in the plumage or the fur of their host. We present the results of a phylogenetic analysis of the Hippoboscoidea based on molecular markers. DNA was sequenced from portions of the nuclear ribosomal gene 28S as well as the nuclear protein coding gene CAD. More than 30 taxa were included representing over 15 genera. Results are presented from both separate and combined analyses using parsimony and Bayesian methods. We discuss the implications for the family-level classification of the Hippoboscoidea as well as the placement of the superfamily within the Calyptratae.

Cladistic analysis of the subtribe Catopina (Coleoptera: Leiodidae) from continental Asia

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The cladistic analysis of the genera of the subtribe Catopina (Insecta: Coleoptera: Leiodidae: Cholevinae) is presented. The analysis is focused on external morphology of adults, including detailed study of characters on male and female terminal abdominal segments and genitalia. The following genera are covered in the study: *Catopodes* Portevin, *Catops* Paykull (with several species groups), *Cholevinus* Reitter, *Dzungarites* Jeannel, *Himalops* Perreau, *Mesocatops* Szymczakowski, *Rybinskiella* Reitter and *Scioepoides* Hatch, with representants of the sister subtribe Cholevina selected as outgroup.

Monophyly of the Papilionoidea (Lepidoptera) families based on the male genitalia

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Recently monophyly of families of the Papilionoidea are established based on morphological and molecular data. However, autapomorphic features of the male genitalia supporting monophyly of butterfly families are rather poorly investigated. In this paper, I worked on this problem. The results are as follows, and all the features stated below are considered apomorphic in the Papilionoidea. The Papilionidae are supported by the presence of the epicosta, which is a detached basal part of the costa of valva. The Pieridae are supported by the presence of the Valvenansatz, which is a prolongation from the laterodistal portion of tegumen, and is articulated with basal portion of costa. The Nymphalidae including Satyrinae and Morphinae are supported by the presence of the appendix angularis, which is the appendicular process for the costa of valva, and is projected from the tegumen close to the vinculum. The Danaidae are supported by the transformation of the scaphium (10th abdominal dorsum) into a pair of setiferous lobes. The Libytheidae are supported by the peculiar prolongation of a pair of sclerotic bands from the tegumen connecting the juxta. The Lycaenidae including Riodininae are supported by the presence of a pair of falces, which are main organ connecting the female genitalia.

Parthenogenetic species in the aphid genus *Colopha* (Aphididae: Eriosomatinae) as a relic

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It is widely conceived that parthenogenetic species are exceptional and difficult to last for a long time in animals. This study tested the possibility of long-term persistence of an aphid species by means of parthenogenesis (relic hypothesis). Aphids of the Eriosomatinae are typically associated with primary and secondary host plants and alternate host plants seasonally. Sexual reproduction takes place on the primary host plant. However, some species are not host-alternating and wholly parthenogenetic on the secondary host plant. The genus *Colopha* is a small aphid group represented by three sexual and three asexual species. The sexual species are associated with two *Ulmus* species that are most closely related and are distributed disjunctively in Europe and eastern North America. In East Asia, an asexual species, *Colopha kansugei*, is distributed widely on the secondary host. We tested whether the relic hypothesis is applicable to *C. kansugei* by constructing molecular phylogeny. A high rate of substitution (3.4% at the maximum) was found in the mitochondrial COI sequence between local populations. Evidence available suggests that *Colopha kansugei* has persisted on the secondary host plant through parthenogenesis as a relic, probably since the local extinction of the primary host.

Computerized checklist and database on Peruvian Chrysomelidae (Coleoptera): Alticinae and Galerucinae

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The family Chrysomelidae *s.l.* (Coleoptera) is one of the most diverse in the Neotropics and throughout the World Tropical Regions. Comprising close to 1000 nominal genera, the traditional subfamilies Alticinae and Galerucinae (now both included in Galerucinae) constitute one of the so-called "Hiperdiverse" groups of living organisms. Circa 200 genera of traditional Alticinae are regarded as endemic for the Neotropical realm. Chrysomelidae importance stems not only from their high diversity and ecosystem services, but also from a number of important pest species in the country and elsewhere. Currently, no checklist of Chrysomelidae is available for Peru or neighbour countries. Latest published listing for Peru dates back to Blackwelder outdated compilation. Here, I present the preliminary results of a computerized taxonomic checklist of traditional Galerucinae and Alticinae from Peru, based on literature and species records of autoritatively identified material housed in research entomological collections in Peru. Data from both literature and individual specimens have been incorporated into a custom-made computer database, which includes taxonomic and geo-referenced distribution data. Database design was based on ASC information model and followed TSWG recommended standards. Data contained in the database will be made accesible through the web, following GBIF data access and interoperability proposal.

New findings on Agyrtodini, with adult and larval descriptions (Coleoptera: Leiodidae)

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The basal leiodid subfamily Camiarinae comprises the tribes Agyrtodini, Neopelatopini, and Camiarini, united by plesiomorphic characters alone. This weakly-supported subfamily contains the most primitive leiodids, and includes many taxa of use for polarizing evolutionary trends within Staphylinoidea. Sadly, Camiarinae is a miasma of unresolved tribal relationships and poorly-known genera with many undescribed species. Although agyrtodines such as *Agyrtodes* and *Zearagytodes* are collected with considerable ease and frequency in Australia and New Zealand, their scientific utility is severely constrained by taxonomic obscurity and a paucity of reliable keys. As a first step towards a better understanding of relationships within Leiodidae, a revision of the genus *Agyrtodes* (the most speciose genus within Camiarinae) is proposed. Keys are provided to adults of *Agyrtodes* and to genera of Agyrtodini. Larval forms are newly described for several agyrtodine species and surveyed for phylogenetically significant morphological characters .

New Zealand Carabidae – a revision of the *Mecodema curvidens* species complex

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Worldwide the Carabidae (ground beetles) family is comprised of 25,000-50,000 species, within 6 subfamilies and 85 tribes. In New Zealand the carabid fauna is small compared to warmer regions. However, several ancient tribes have reached their greatest diversity and endemism is high at both genera (58%) and species (92%) levels. One tribe, the Broscini is represented by 5 genera, of which *Mecodema* is the most species-rich with 74 taxa divided into 8 species groups. My research will focus on a revision of the *Mecodema curvidens* species group, which has five species listed as endangered. This complex has a unique distribution in coastal forests of the North Island's northeast coast, from Cape Reinga to the East Cape and on four offshore island groups. Both morphological characters (external and genitalia) and molecular analysis will be used to resolve taxonomic confusion of the 5 currently described species and identify a possible 7-9 new species. Population distributions will be looked at through pitfall trapping within suitable habitats along the recognized distribution. This research will increase the overall knowledge of *Mecodema* beetles in New Zealand and give more insight into the distribution and species diversity of the *curvidens* group, allowing conservation groups to concentrate management on rare or endangered species.

Phylogenetic relationships in the Swallowtail Butterflies of the tribe Troidini (Papilionidae) based on COI, COII, and Ef1a Genes

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Butterflies of the tribe Troidini (Papilionidae, Papilionini) are predominantly tropical, with three genera present in the New World: *Battus*, *Euryades*, and *Parides*. They are often regarded as classic examples of coevolution with their larval host plants (Aristolochiaceae). There are three different hypotheses of Troidini phylogeny, all based primarily on morphology. The central aim of this work is to obtain a phylogenetic hypothesis for this group based on molecular characters. We have sequenced three genes, COI and COII (mitochondrial), and EF-1α (nuclear), in 20 species of Troidini. Phylogenetic analyses were performed with PAUP, using Maximum Parsimony and Maximum Likelihood on the full data set (3285 nucleotides; 890 informative sites). Parsimony searches gave two most parsimonious trees, with 3400 steps (CI=0.456, RI=0.683). The monophyly of the Troidini clade was supported by bootstrapping with 92% confidence, still more likely was the *Parides* clade (98%). The genus *Battus* appeared as basal, followed by *Euryades, Parides, Pachliopta* and *Troides+Atrophaneura*. The maximum likelihood analysis for the combined data set under the GTR+I+G model, revealed a single tree with a score of –ln *L*=21564.053, very similar to those obtained by MP. Separate analyses showed that there is significant congruence among mitochondrial and nuclear genes (ILD test, p=0.746).

Molecular phylogeny of Larentiinae and Ennominae (Lepidoptera: Geometridae)

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Geometridae is a family with more than 20 000 described species. In this study, we focus on the subfamilies of Larentiinae and Ennominae, and use nuclear DNA markers in order to resolve some of the ambiguities of the proposed phylogenetic relationships. Further, we are interested in how many independent changes from winged to wingless females there have occurred. Portions of nuclear DNA genes, 28S rRNA and EF-1α, were sequenced from 36 species. Sixteen Geometridae and six outgroup sequences published elsewhere were combined with our data. Direct optimisation was used to infer a phylogenetic hypothesis from the combined sequence data set. Our preliminary results show that Larentiinae form monophyletic group, but Ennominae is paraphyletic. In both subfamilies traditional tribes are not fully supported. Furthermore, the current data suggests that in both subfamilies female winglessness has evolved independently more than once. Future plans of this work include sequencing one more gene fragment and increasing the number of taxa studied.

A new genus and the larvae of Tanygnathinina (Coleoptera: Staphylinidae: Staphylinini)

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Until recently, the subtribe Tanygnathinina (Staphylinidae: Staphylininae: Staphylinini) contained only the single genus *Atanygnathus* (about 50 described species; mostly tropical; predominantly hygrophilous). Morphologically, *Atanygnathus* is a rather peculiar taxon of Staphylinidae with substantial controversies about its taxonomic placement within the family. Recently a less specialized new genus (including its larvae) closely related to *Atanygnathus*, and larvae of *Atanygnathus* have been found. Detailed morphological examination of the adults and larvae of both genera revealed new synapomorphies of Tanygnathinina with the rest of Staphylinini. Several very interesting autapomorphies of Tanygnathinina and *Atanygnathus* have now been found, too, especially in larvae, and some earlier interpretations of particular adult character states are put in doubt. The new findings support the recently published idea about the isolated position of Tanygnathinina within the tribe Staphylinini. Probably, Tanygnathinina is a highly derived lineage of Quediina. At the moment, however, meaningful testing of this hypothesis is impossible because the composition of Quediina is not properly understood on a world basis, and larvae of Staphylinini are still inadequately known.

The Agathidinae (Hymenoptera: Braconidae) of Australia; parasitoids of lepidopteran larvae.

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One of the largest parasitoid wasp families is the Braconidae (Hymenoptera) with nearly 15,000 valid species world-wide, and possibly three to four times this many species waiting description. The Braconidae are well represented in Australia with 29 of the 34 subfamilies recognised occurring here. However, the Australian fauna has received little attention. This project focuses on the systematics of the Agathidinae in Australia. This subfamily is a relatively speciose group with over 1200 described species in over 50 genera worldwide. Little is known of the Australian fauna with only 36 species in eight genera described to date. Agathidine taxa are solitary endoparasitoids that parasitise lepidopteran larvae, most commonly concealed larvae. The distributions of described agathidines show they are diverse in a wide range of climatic zones from tropical/subtropical regions, to more temperate and arid environments. The Australian continent encompasses a wide range of climatic regions, providing a myriad of habitats into which agathidines have diversified. Preliminary investigation has discovered many new species. The most dramatic example is for the cosmopolitan genus *Bassus* Fabricius which is presently represented in Australia by only one introduced species. However, over 25 new taxa have already been discovered.

Systematic study of the *Diplodoma* genus group (Lepidoptera, Psychidae)

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In the family Psychidae, body structures of adult females are reduced in various degrees, and morphology of adult males and biology of both sexes associated with the female body reduction show various specializations. Therefore this family is suited to evolutionary study. However, as the adult morphology is so diversified that no stable taxonomic system has been established, and intrageneric phylogenetic relationships in the family are poorly known. In this paper we studied on morphology and ethology of the monophyletic *Diplodoma* group that includes two types of genera, one with winged and legged females, the other with wingless and legged females, and analyzed phylogenetic relationships among the genera. As the result, this group is divided into five clades. One clade is supported by the presence of apomorphic fultura inferior in the male genitalia, and includes *Diplodoma*, *Narycia* and *Paranarychia* with winged females and *Dahlica* and its allies with wingless females. The reduction of female wings appeared also in other three clades. Therefore, the wing reduction occurred many times in different clades in the group, and it suggests that reductions of female body structures appeared independently in various groups in this family. A preliminary work on molecular systematics of the group is also mentioned.

Geographical variation of the male genitalia of *Sciara humeralis* Zetterstedt, 1851 (Diptera, Sciaridae)

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We have collected 8 species of *Sciara* from Japan: *S. thoracica*, *S. hemerobioides*, *S. helvola*, *S. multispinulosa*, *S. humeralis*, *S. melanostyla*, *Sciara* sp.1 (undescribed sp.), *Sciara* sp. 2 (undescribed sp.) (Sutou, unpublished).
One of them, *S. humeralis* is known to be widely distributed in the Holoarctic region, and also from the Oriental region. Menzel and Mohrig (1991, 2000) pointed out the morphological variation of the male genitalia of this species, and categorized it into 3 types based on the materials from Europe, Taiwan, and USA. We call them Palaearctic type, Oriental type, and Nearctic type, respectively. So far, Oriental type has been recorded from Taiwan, southern China (Guizhou Province), and northern Vietnam. We recorded many specimens of Palaearctic type from mainland of Japan (Akita, Iwate, Tokyo, Kagoshima), and Oriental type from southernmost islands in Japan (Ishigaki Is. and Iriomote Is.). Based on these materials, we estimated that the distributional borderline between Palaearctic type and Oriental type lies within southern islands of Japan.

Phylogeny of the bee flies (Diptera: Bombyliidae)

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Bombyliidae, or bee flies, rank among the largest and most diverse families of Diptera. This striking diversity contributes to the complexity of determining their evolutionary relationships using comparative morphology alone. Only a small number of studies, however, have relied on molecular data to reconstruct phylogenetic relationships within the Bombyliidae. The most recent comprehensive phylogenetic analysis of bombyliid subfamilial relationships was based on morphological characters. Our current molecular study provides additional data to confirm, test, and further resolve higher-level classification within the Bombyliidae. To do this we sequenced large fragments of CAD, a protein encoding gene, and the 28S ribosomal DNA for more than 35 taxa, representing 12 of the 15 currently recognized subfamilies, along with several outgroups. Based on phylogenetic analyses of these data using parsimony and Bayesian methods, we assess support for the monophyly of currently recognized subfamilies and the position of various genera within the family. An analysis of congruence between the DNA sequence data sets and previously compiled morphological data is also presented. Evidence in support of alternative sister groups for the Bombyliidae is discussed within the broader context of relationships among asiloid Diptera.

The redefinition of the ejaculatory reservoir *s. str.* in Pentatomomorpha (Hemiptera: Heteroptera)

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Singh-Pruthi (1925) defined the ejaculatory reservoir as "an expansion of the ejaculatory duct at the base of the vesica [= aedeagus] present in Heteroptera." After carefully examining the external male genitalia of Dinidoridae, Scutelleridae, Tessaratomidae, and Cydnidae, we found that Singh-Pruthi's definition could not be satisfactorily applied to the above families. Therefore, it is necessary to redefine the ejaculatory reservoir in the Pentatomomorpha. We found that the structure protrudes from the dorsobasal portion of the expansion of the ejaculatory duct within the apical half of the aedeagus. We redefine that part of the structure outside the base of the aedeagus as the "ejaculatory reservoir" and the other part within the aedeagus, including the expanded ejaculatory duct, as the "reservoir lumen." There are three differences between our and Singh-Pruthi's definition of the ejaculatory reservoir. (1) The position (an evolutionary event) within the middle of the aedeagus vs. at the base of the aedeagus, (2) The nature of the structure protruding from the expanded ejaculatory duct vs. an expansion of the ejaculatory duct, and (3) The presence in only Pentatomomorpha vs. in Heteroptera.

Phylogeny and classification of the Iceryine Margarodids (Hemiptera: Coccoidea)

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This study examines the taxonomy and evolutionary relationships of selected members of the tribe Iceryini within the scale insect family Margarodidae. This tribe is monophyletic and belongs to the subfamily Monophlebinae. Relationships among iceryine genera and species are largely unknown. The only phylogenetic studies of margarodids are a cladistic analysis of morphology including three genera of Iceryiniand an unpublished preliminary molecular phylogeny. The subfamily Monophlebinae consists of five tribes, of which the Iceryini is the largest and best known because many species are important pests of commercial agriculture across the world. The six genera in this tribe are *Auloicerya* Morrison (2 species), *Crypticerya* Cockerell (15 species), *Echinicerya* Morrison (1 species), *Gueriniella* Fernald (2 species), *Icerya* Signoret (40 species) and *Steatococcus* Ferris (14 species). This poster presents a preliminary molecular analysis of the iceryine tribe and its outgroups with species from the Americas, Australia, New Caledonia, Hong Kong and Thailand. These molecular data are from the mitochondrial gene cytochrome oxidase subunit I (COI) and a nuclear gene from the D2 expansion region of the large subunit ribosomal DNA gene (28S).

Phylogeny of fossil and extant Sciaroidea (Diptera: Bibionomorpha)

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The phylogeny of the main groups of Sciaroidea, including the fossil Antefungivoridae†, Archizelmiridae†, Mesosciophilidae†, Pleciofungivoridae, Pleciomimidae†, Protopleciidae† and Bolitophilidae: Mangasinae†, and an extant new taxon, was studied by parsimony analysis. Two cladistic analyses of 78 morphological characters from adults were made. One analysis, with forty-one extant taxa in the ingroup and the other, with twelve fossil taxa in addition, produced both two most parsimonious cladograms. The phylogenetic hypotheses obtained differed from each other, and partly largely from previous ones although the most traditionally recognized groups appeared monophyletic, including the speciose Cecidomyiidae and Sciaridae. Cecidomyiidae (fossil analysis) or Keroplatidae-Ditomyiidae (extant analysis) appeared as the sister group of the rest of the Sciaroidea. According to the present solution, we propose to emend the current Sciaridae to include five subfamilies: Archizelmirinae† stat. n., Rangomaraminae stat. n., Sciarinae, Subfam. n.1 and Subfam. n.2. A new genus from Namibia is described, and, according to the phylogenetic analysis, placed into Sciaridae, as the type-genus of one of the new subfamilies. As the sister group of Sciaridae in its new concept appeared the Mycetophilidae group, in the extant analysis including Mycetophilidae, Manotidae, Lygistorrhinidae, *Pterogymnus* and *Sciaropota*, in the fossil analysis also Mesosciophilidae† and the *Ohakunea* group (*Ohakunea* + *Colonomyia*).

Systematics, biogeography and conservation in the *sari* group of Old World *Eurema*

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The *sari* group, the largest group of the genus *Eurema* occurring in the Old World, has its distributional center in the Oriental Region and is a distinct monophyletic group consisting of 18 species and 4 subgenera (Yata, 1989). Since the group consists of many rare and local species inhabiting forests and shows remarkable allopatric speciation on Southeast Asian Islands, it is suitable to discuss the phylogeny and biogeography of the butterfly. The phylogenetic relationships within the subgenus *Terias* including the *sari* group were inferred in a revision of the genus *Eurema* (Yata, 1989). This revision, however, was mainly based on morphological characters and DNA information not used. In the present study, the phylogenetic relationships among 12 *Eurema* species including this group were inferred using the base sequence of COI region of mitochondrial DNA (mtDNA). The monophyly within three groups, *hecabe - alitha*, *nicevillei – tilaha - lombokiana* and *novapallida - lacteola*, was distinct and well coincident with that from morphology, respectively, but not for *hiurai* from the Philippines including the subgroup *lacteola* with *novapallida - lacteola*. Based on these results were discussed the systematics / biogeography of these groups and the meaning of their habitat for conservation biology.

Preliminary phylogenetic analysis of Musotiminae (Lepidoptera, Crambidae) and the implications on biological control of weedy ferns

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The crambid Musotiminae has 24 genera recognized from around the world, most of which are obviously not monophyletic, and many species are undescribed. We seek to address the following aspects of musotimine systematics: (1) what are the relationships of this subfamily to other crambid subfamilies; (2) what are the synapomorphies of this subfamily; (3) what can new adult morphological characters tell us about the relationships within the group. We show that several 'clades' within the subfamily can be defined based on some putative synapomorphies, but their internal relationships are blurred because information of immature stages is still scarce. Fern-feeding behaviour of the musotimines from different parts of the world has been discovered in the past two years, and thus these moths may have potential for controlling the weedy ferns. We mapped the known host fern families on to a preliminary genus-level phylogeny of Musotiminae and found no evidence of co-phylogenesis between the fern families and moth genera though there is a tendency for closely related species and genera to utilize the same fern family. However, some fern families such as Schizaeaceae have been colonized by unrelated lineages of musotimines. We expect that more exploration for biological control agents will result in an improved picture of the fern-musotimine relationships.

Evolution of the Australian endemic Geometridae

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The Australian endemic Geometridae form a large part of the Australian indigenous macromoth fauna. Most of these species belong to the tribe Nacophorini. New molecular and morphological evidence from all stages of the life cycle enabled the characterisation of this group in a phylogenetic framework. The Tasmanian Archiearinae was well separated from the Holarctic fauna on both molecular and morphological evidence. The alpine taxa of this group are most likely an endemic clade closely related to the Nacophorini, but may have affinities with the Neotropical Archiearinae. The proposed phylogeny from this work provides a context for the interpretation of the evolution of key characters such as the presence or absence of extra prolegs, host-plant relationships and the biogeography of the Nacophorini. It is apparent from this phylogenetic evidence that the development of extra prolegs in geometrids may be a labile character, subject to independent reversals in several geometrid lineages. The relationship of the Tasmanian Archiearinae with the Nacophorini is very well-resolved and provides an interesting case-study in insect-host plant relationships. The phylogeny of the global Nacophorini is explained by Gondwanan fragmentation. Furthermore apparent close relationships between the American Nacophorini and another Nearctic ennomine tribe can be explained by recent dispersal of Neotropical taxa to North America.

Phylogeny of the Australian Geometridae

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The Australian endemic Ennominae, frequently postulated as a possible primitive group within the Geometridae, was described and characterised using material from all stages of the life cycle. These descriptions were used as a basis for the construction of a morphological phylogeny for the Australian Ennominae in the context of the world fauna. Sequences from two nuclear gene fragments were also used to construct a molecular phylogeny in order to more confidently propose a robust evolutionary pathway for the Australian Geometridae. Phylogenetic trees obtained from all data sets recovered almost identical, supported relationships. The major findings from these analyses were as follows (a) The Epanidae as sister group to the Geometridae; (b) The Larentiinae as basal within the Geometridae; the Sterrhinae as the penultimate basal group, except that in the morphological analysis this relationship is reversed so that the Sterrhinae are basal; (c) The Oenochrominae s. str. as sister group to the Geometrinae; (d) The Ennominae as not monophyletic; (e) The Tasmanian Archiearinae as misplaced in the Archiearinae and with close affinities to the Australian Nacophorini; (f) The Australian Nacophorini as not monophyletic. These results were at odds with traditionally held beliefs on the origins of the Geometridae.

Phylogeny of the Gelechioidea: Evolution of larval food substrate use

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The phylogeny of the lepidopteran superfamily Gelechioidea was estimated using 193 characters derived from larval, pupal and adult morphology and larval ecology. 143 species represented the diversity of the putative Gelechioidea in the analysis, and further 13 species representing three monotrysian and eight other ditrysian families were included to test the monophyly of the Gelechioidea. The Gelechioidea was nested within Apoditrysia instead of being its sister group as previously suggested. The well-resolved cladogram suggested a largely different concept of interrelationships of constituent gelechioid families as compared to other studies. The Gelechioidea was basally divided into two main lineages, herbivory as the ancestral larval feeding strategy of one, and, depending on character optimization, either saprophagy or herbivory of the other. Therefore it is not possible to unambiguously determine which of these traits is the ancestral one in the Gelechioidea. It is also sometimes questionable whether such a classification of the food substrate can be applied, .e.g. if the larva uses dry seeds and other dry vegetable matter. Detritivory/fungivory may have evolved only twice within Gelechioidea, though the evolution of larval food substrate use has been quite dynamic with frequent reversals.

Systematics of parasitoids (Hymenoptera: Chalcidoidea) of the Emerald Ash Borer ,*Agrilus plannipennis* (Buprestidae).

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In May 2002, an unknown species of beetle was reported by researchers to be attacking and killing at least 3 of the 4 native species of ash trees (*Fraxinus spp.*) in southeastern Michigan. The species was subsequently identified as the Emerald Ash Borer (EAB), *Agrilus planipennis* (also known as *Agrilus marcopoli*) on July 08, 2002. Extensive mortality of ash trees attributed to EAB has been reported in the Detroit area of Michigan and it appears to be a damaging pest to trees growing in both urban and forested sites. Researchers at the U.S. Forest Service and Michigan State University have reared two parasitoids (Hymenoptera: Chalcidoidea) of EAB from a study plot in Livonia, Michigan, USA. We received representative specimens of these parasitoids for identification and determined them to be *Balcha* sp. (Eupelmidae) and *Pediobius* sp. (Eulophidae). Both species are believed to be undescribed. The *Pediobius* species is a parasitoid of ash borer eggs while the *Balcha* attacks *Agrilus* larvae. Foreign exploration is continuing to try to find additional parasitoids. Details of the systematics and biology are given.

An interactive key to *Liriomyza* parasitoids in South East Asia

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Invasive *Liriomyza* species are a severe pest in South East Asia, affecting a variety of vegetable and ornamental crops. An interactive, multiple entry key, using Lucid software, has been developed for identification of indigenous parasitoids of SE Asian vegetable leafminers. This key will support activities aimed at sustainable management of these pests. The key includes 20 species from 4 families (Braconidae, Eucoilidae, Eulophidae, Pteromalidae). Species pages are provided for all species, which give information on: classification, diagnostic characters, distribution, hosts, biology, comments and references. Additional information pages are presented for each family, subfamily, and genus. A tutorial section is included to facilitate the use of the key.

Non-destructive investigation of the internal anatomy of insects utilising high-resolution X-ray computed tomography

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The investigation of the internal anatomy of insects involves very time consuming processes of preparation and usually results in the destruction of the specimen. Nevertheless, detailed knowledge of insect anatomy is indispensable for many research areas like phylogenetic investigations, parasitology, etc. During recent years the authors optimised the application of high-resolution X-ray computed tomography (µCT) for the study of the anatomy of small insects. This method has a number of advantages over methods like serial-sectioning and free-hand preparation: Due to the very simple preparation and the fast and artefact-free data sampling it is possible to investigate a large number of specimens in very short time. This, e.g., allows for studies of intra- and inter-specific variation of internal structures. Further more, the specimen remains intact, which is very important for valuable specimens like holotypes. The data sets are easy to handle and it is possible to produce virtual sections in any desired cutting plane from one data set. Also the preparation of virtual (and real) 3D models is straightforward, since there are no deformed or missing sections. The 3D models are excellent tools for teaching insect anatomy. Here the preparation of specimens, the data-acquisition, and some examples of data handling, interpretation and presentation are demonstrated.

Tracking specimens, taxonomic nomenclature, & literature in Mandala

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Mandala is a Windows and Macintosh-compatible data management system designed in FileMaker Pro to support four major realms of data acquisition and management for systematics and biodiversity studies: specimens, literature, taxonomic names, and illustrations. Development began in 1995 as part of a US National Science Foundation PEET project on the fly family, Therevidae, and has progressed into the current feature-rich system that is freely available upon request. Mandala details the complex history of taxonomic names; exports data for phenological plots, specimens examined lists, and distribution maps; links to GenBank records; tracks museum loans and batches of organisms; catalogues images and illustrations; stores and links literature to appropriate taxonomic names, specimens, illustrations, and collecting localities; and records numerous details about individual specimens including a literal transcription of the label(s), enhanced locality and collecting event data as interpreted from the label(s), taxonomic determination history, physical condition of the specimen and how it has been preserved, ecological/biological associations with other collected specimens or other taxa associated but not collected, and type designation. Mandala also features context sensitive help, electronic problem management, and tracks structural changes made to Mandala's architecture. Mandala is a powerful tool packed with features waiting to be exploited.

Pipilika: an identification and information system for the ant fauna of India

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Pipilika is a digital key for identification of ants of India featuring a minimum technical language is provided to assist the identification of all subfamilies and genera. The sources of information for the development of this key are diverse, ranging from C.T. Bingham's work on the Indian ant fauna, to works and collections of various myrmecologists and individual researchers from India and overseas. This is the first attempt to provide a digital checklist for the fascinating and diverse Indian ant fauna. The user-friendly software is an innovative and comprehensive program that enables one to make accurate identification of ants. The most important diagnostic attributes (characters) are used to describe a taxon. Hence, each taxon has a unique combination of characters assigned to it. It prompts the user to answer a series of questions about the specimen that needs to be identified. These questions have choices, which are made on screen from text. Then, through a process of selection, the key interprets the user's answers to reach an identification of the specimen. Each subfamily and genus includes information on its biology, identification and distribution, illustrations and a summary of publications dealing with the subfamily and genus.

An interactive key to sharpshooter genera of the tribe Proconiini (Hemiptera: Cicadellidae)

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The New World Proconiini comprises approximately 350 species in 56 genera, all potential vectors of the bacterium *Xyllela fastidiosa* causing diseases of over 20 economically important plant species. The last taxonomic revision of the tribe (Young 1968: *Bull. U.S. Nat. Mus.* 261) contained a dichotomous key to genera. Since this revision, one additional genus has been described, two are in manuscript format, and at least three should be transferred to this tribe. Several steps in Young's key emphasize characters that are subjective (e.g., claval veins fused almost throughout their length), variable (e.g., *Pseudophera* species smaller than 16 mm will key to *Homalodisca*), or from the male genitalia of difficult study by non-specialists. An Internet-based interactive generic key will update and facilitate the identification of this economically important leafhopper group by enabling users to choose characters in any order and providing illustrations explaining each character states. A list of all species, photographs of over 100 species, and bibliography are also included. The key was developed using the software package 3I (Internet-accessible Interactive Identification) composed of a MS Access database and ASP pages for Internet publishing developed by the junior author. The key is still under construction and hosted at http://ctap.inhs.uiuc.edu/takiya.

Acridoidea fauna of Brazillian Amazonia

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The superfamilly Acridoidea (Orthoptera) is considered of economical importance because its species may cause enormous damages to the cultivations and sometimes destruct entire crops. The knowledge of this species, its taxonomy and geographical distribution are of great importance to the science. With the purpose to increase the knowledge of this superfamilly, a list of the species from the Brazilian Amazonia was elaborated. To complement this work, the taxonomy and geographical distribution of the species are bring up to date, as well as the location of the types and principal bibliographical records. Until now the list comprises six families, twelve subfamilies, one hundred and sixty-four genera and five hundreds and forty-nine species. The numbers show the great diversity of Acridoidea in this area, one of the most riches in grasshoppers. A major part of the species listed are deposited in the collection of Museu Nacional – UFRJ and are originated from collects realized by staff of this museum, others were donated from others collections or were included based on information from bibliographical records.

A review of the Southern African Acroceridae (Diptera) with particular reference to the endemic genus *Psilodera* Gray

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The recognized Southern African fauna of Acroceridae is reviewed (10 genera – *Astomella*, *Corononcodes*, *Pterodontia*, *Stenopialea*, *Africaterphis*, *Thyllis*, *Acrocera*, *Ogcodes*, *Psilodera*, *Sabroskya*); their life history discussed and gaps in the literature highlighted for future investigation. One of the two endemic genera, *Psilodera* Gray, is assessed in more depth. The nine named species of *Psilodera* are large, conspicuous flies that appear to be the only long-proboscid acrocerids in Africa. Two strongly defined species-groups (*fasciata* group with five species and *bipunctata* group with four species) are suggested from this ecologically interesting genus, using characters such as the genitalia, not fully explored previously. The *fasciata* group includes species with a dark thorax and abdomen, and minutely branched pile, while the *bipunctata* species have a patterned thorax and abdomen and simple pile. The more clearly understood pollination activities of adults are given. They pollinate three endemic *Plectranthus* species (*P. ciliatus, P. hilliardiae* and *P. zuluensis*) as well as species of *Impatiens*, *Gladiolus* and *Aeolbnthus*. The known distribution of the *fasciata* group includes all of South Africa (the arid Northern Cape excepted) and the esatern highlands of Zimbabwe. The *bipunctata* group is found in the southern and eastern coastal provinces of South Africa.

Atlas of the butterflies of the Iberian peninsular and Balearic Islands

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The Atlas is the first referred to the distribution of the butterfly species of the Iberian Peninsula and Balearic Islands. The maps use the UTM system with a 10 x 10 km grid. The Atlas has been produced by research teams from Spain and Portugal have taken part of it. In the making of the Atlas we started compiling a database (ATLAMAR1) with the distribution data on Iberian butterflies. Each record in the database contained at least four fields: 1) name of the species, 2) name of the locality in which it had been recorded 3) reference to the UTM grid system using 10 km squares and 4) identification of the source from which the information came (publication, collection, etc.). For ATLAMAR 1 we have gathered a total of 292000 records. Data come from three different sources: 1) bibliographic information, 2) data from collections and 3) unpublished data from the authors and collaborators. Bibliographic information represents approximately 73% of the records, and is based on the revisión of more than 1500 references in journals and books. These data gather information on at least 421500 butterfly specimens. We estimate that a 98% of the relevant bibliographic information has been compiled for the project. Collections represent around 37000 records, and another 45000 records come from unpublished data.

A complete beetle fauna on the web: Mauritius as a model case

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Mauritius is a tropical, mountainous island in the Indian Ocean of 1865 km^2. Its beetle fauna comprises around 1020 species and has been covered by a comprehensive catalogue of Vinson (1956-1962) and recently by a synoptic list by Yves Gomy (2000). Keys and illustrations are available for only a few small groups. Jean Vinson deposited 1480 specimens of his beetle collection in The Natural History Museum, London, in 1972. On the basis of these specimens and additional material to be collected within the Darwin Initiative Programme 'Rediscovering neglected insects of Mauritius: building in-country capacity', we compile a web-based catalogue of the complete Mauritian beetle fauna, illustrated by colour photographs of each available species, and accompanied by references on economic importance and ecology of the species. This will be the first web catalogue of a complete beetle fauna on an up-to-date taxonomic basis and might serve as a model case for similar, larger-scale projects.

Taxonomic survey of the South-East Asian Ethmiidae (Lepidoptera)

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The genus *Ethmia* is cosmopolitan, more than 40 species were shown from the south-eastern Palaearctic. The Ethmiid fauna of China is known as the richest within this region with 33 formerly recorded taxa; by comparison 11 have been recorded from India, 6 from Nepal, 9 from Japan, only three from Korea, 7 from the Philippines and 14 from Australia. In the last few years several Hungarian lepidopterists worked intensively in different regions of South East Asia, including Taiwan, Vietnam, Pakistan, South Korea, and Thailand. Due to the effective methods used in these expeditions, a large material of Ethmiinae containing more than 300 individuals has been collected during the last five years. As a result of the treatment of this material and of the collections of the British Museum (Natural History), London; Zoologische Staatssammlung München; Staatliches Museum für Tierkunde, Dresden; Landesmuseum für Naturkunde, Karlsruhe; Witt Museum, München; Taiwan Forestry Research Institute, Taipei; Center for Insects Systematics, Kangwon University, Chuncheon, Korea; National Institute of Agricultural Science and Technology, Suwon, Korea; the Hungarian Natural History Museum, Budapest, as well as the survey of the literature data, further 11 *Ethmia* species were described as new to science, with taxonomic comments and corrections. The Ethmiid fauna of the region is, however, still not well-explored and several taxonomic novelties are expected from the continuation of the research.

A faunistic analysis of Chinese phorid flies (Diptera: Phoridae)

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The Phoridae fauna in China is zoogeographically heterogeneous due to the geological diversity of this region. On basis of our collection, the Chinese phorid flies are grouped into three principal complexes: Palearctic, Oriental and Boundary complexes. Species in Northeastern, Northern China and Xinjiang-Mongolia region are included in Palearctic complex, which mainly belong to Palearctic elements. The species composition in Xinjiang-Mongolia region is more similar to central Europe type than that in Northeastern- and Northern China. Species in Centro-China, South-China and West-southern China area belong to Oriental complex. This complex is rich in species. More than 50% of record species of China belong to the complex. Some of species spread to Palearctic region, for example, Diplonevra peregrina (Wiedemann). The Qing-Zhang plateau is the boundary of Oriental and Palearctic region. More than 3000m-altitude constitutes a special ecological environment. Species in the region are morphologically distinguished from others and comprise a special complex.

An overview of the phylogeny of the Brentoid Complex (Col.: Curculionoidea)

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The so-called Brentoid Complex includes a group of taxa which are thought to be evolutionarily intermediate between the "lower" weevils (Nemonychidae to Attelabidae) and the "higher" weevils (Brachyceridae to Curculionidae). The complex seems to have originated in Gondwana. The status of the included groups is much disputed: Ithyceridae, Brentidae (+ Cyladinae), Eurhynchidae, Apionidae (+ Antliarhininae) and Nanophyidae. Carina has also been related to this group. While most of these groups seem to be clearly monophyletic on morphological grounds, their relationships inside this complex or even whether this complex is monophyletic are not clear. Current analyses of morphology and DNA sequences are contradictory. Wanat (2001), after a thorough cladistic analysis of morphological data, concluded that the families Brentidae (including Cyladinae), Caridae, Nanophyidae, Eurhynchidae and Apionidae (incl. Antliarhininae) are well supported. Ithyceridae have been placed (Marvaldi & Morrone 2000) as a subfamily of Curculionidae. Oberprieler (2000) and Marvaldi (2002) exclude the Carinae from the Brentidae. Nanophyidae were related by Zimmerman (1993) to the most primitive of the "higher" Curculionoidea (his Heteromorphi). However, promising preliminary molecular data by the senior author relate Caridae to Nanophyidae as part of this complex. The internal phylogeny of the diverse family Apionidae is also matter of debate.

Unity of Psychodomorpha? The position of Trichoceridae & Anisopodidae in the system of the Diptera based on thorax morphology

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The Trichoceridae have been traditionally placed in the Tipulomorpha, together with Tipulidae, Limoniidae, and Cylinotomidae. Most authors included the Anisopodidae in the Bibionomorpha. The position of both families was challenged by studies using immature morphology. From the immature features perspective, Trichoceridae and Anisopodidae fit with Psychodidae, Perisommatidae, Canthyloscelidae, and Scatopsidae into the Psychodomorpha. A detailed study of thorax morphology corroborates that Trichoceridae stands apart from the Tipulomorpha, nested into the Psychodomorpha. Synapomorphies for all Diptera except Tipulomorpha are shared by Trichoceridae, while apomorphic features shared by the Psychodomorpha families are clearly present in Trichoceridae. The Anisopodidae, however, are obviously plesiomorphic for the thorax features seen in the Psychodomorpha, it additionally shares modified characters with the Bibionomorpha families. Thorax morphology, hence, corroborates that immature information is powerful to develop a phylogenetic system for the Diptera, but at least some of the immature features are clearly subject to homoplasy. Plates of thorax morphology of basal families of Diptera are presented.

Mitochondrial gene rearrangements & the evolution of eusociality in bees

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Mitochondrial DNA sequences data and, more recently, gene order have been widely used as characters to infer phylogenetic relationships. For the subfamily Apinae, phylogenetic relationships have been inferred from morphological and sequence characters, although considerable ambiguity remains. We recently sequenced the mitochondrial genome of *Melipona bicolor* and compared the gene order to *Apis mellifera*. Nine tRNA gene translocations were observed. These translocations identified between two highly eusocial species led us to investigate the mtDNA gene order among Apinae tribes. So far 10 species, representing 8 tribes, have been analyzed for all the tRNA gene positions. Several rearrangements were reported. The very conserved order D-K was present in all species excepted in *Melipona*, which had only the gene D. We expanded our analyses to 5 Meliponini genera and data showed that the presence of D and the absence of K was a very conserved character in this tribe. To better address questions concerning the origin and evolution of social behavior in Apinae, we need to obtain more data about tRNA rearrangements. In addition to investigating mtDNA gene order, we sequenced three mitochondrial genes (16S, COI and CytB). Sequence data support Meliponini and Bombini as sister taxa and a dual origin of eusociality in the Apinae.

Molecular markers in identification: lessons from the Tephritidae

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Exploitation of molecular markers for the identification of tephritid species and populations has been adopted by the quarantine authorities of New Zealand, Australia and North America. The lack of morphological markers for immature life stages associated with the export of fruit and the inappropriateness of morphology for distinguishing populations within a species left managers ill equipped to monitor and control the exotic and cryptic invasion pathways of key high impact horticultural pest species, in particular the medfly and oriental fruit fly. Development of DNA markers, such as polymorphic restriction profiles and microsatellite loci, has overcome this. Now, across laboratories, the identification of around 90 species is supported by DNA markers, several of which can also be traced at the population level. This reflects a growing collaborative effort between molecular and taxonomic researchers and the regulatory authorities. Advances recently have been in a convergence of approaches, preferring to apply novel bioinformatic and statistical analyses of whole sequence data for a few common gene regions. This methodology provides a much more flexible platform on which to continue building identification profiles for additional species and populations and is aimed at superseding the previous disparity in molecular methods.

Phylogeny & homologization of genital sclerites of Neuropterida - arguments between Skylla & Charybdis

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Most recent molecular and previous holomorphological cladistic analyses of Neuropterida coincide in revealing 1) Raphidioptera + (Megaloptera + Neuroptera), and 2) the suborder Nevrorthiformia as adelphotaxon of all other Neuroptera. Conflicts arise with the disruption of the suborder Hemerobiiformia into Sisyridae, Osmylidae, and a clade comprising the remaining Hemerobiiformia, and including the Myrmeleontiformia. Segment nine of male Raphidioptera with tergite, sternite, gonocoxites, styli, and gonapophyses, serves as a model of reference for homologization of male and female terminalia of all three orders. Phylogenetic relevance is obvious, conflicts derive from 1) independent/joint obliterations or modifications of sclerites, 2) parallel apomorphies in derived representatives of diverse taxa (e.g. threadlike penisfilum in certain Coniopterygidae, Berothidae, Rhachiberothidae, Mantispidae), 3) reappearance of serial homologous structures (female gonocoxites and gonapophyses as digitiform processus and flat appendices in segment eight of certain Myrmeleontidae, as wart-like processus and flat circular sclerites in segment seven (!) of certain Berothidae, gonocoxites eight as pseudosternites in certain Nemopteridae, but digitiform processus of tergite 9 as pseudogonocoxites in Rhachiberothidae).

Neighbour-joining goes the distance: a comparison of methods for analyzing DNA barcoding data

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Short DNA sequences, called DNA barcodes, have been shown to provide a highly reliable means of species identification for a diversity of animal taxa. However, there has been disagreement over the appropriateness of currently used methods for analyzing DNA barcoding data. DNA barcoding has relied upon Neighbour-joining (NJ) of Kimura-2-Parameter distances to group sequences based on sequence similarity. This distance-based approach has been highly criticized despite its extremely high success rate in species identification. Here, we compare four methods to test their suitability in barcoding data analysis. We assessed two distance-based methods (Neighbour-joining, Neighbour-net) and two character-state based methods (Maximum likelihood and Bayesian analysis) using three independent datasets. Each method was assessed for: 1) percentage of correct species identifications; 2) speed of analysis. Results showed that NJ was as or more successful (success rate = 95-100%) in correctly identifying species compared to non-distance based methods and also provided the most rapid means of analysis. Given the reliability and speed of analysis, we conclude that this approach provides a highly effective means of species identification through DNA barcoding.

A combined morphological & molecular approach to reconstruct the phylogeny of delphacid planthoppers (Hemiptera: Fulgoromorpha)

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Although the Delphacidae are the largest family of planthoppers (Fulgoromorpha), and include species of major agricultural importance; surprisingly little is known about their phylogeny. The two current competing hypotheses are morphologically based and consider delphacid phylogeny only among the higher taxa (subfamilies, tribes). Here we combine morphology and molecular techniques to reconstruct the phylogeny of delphacids with particular emphasis on the genera of Delphacini, an advanced tribe which comprises 80% of delphacid genera and 60% of species. At present, these analyses are preliminary and based primarily on taxa from Eastern North America. For these analyses, a morphological dataset and four molecular datasets (EF-1a, wingless, 18S rDNA, and 28S rDNA) were generated. Individual datasets did not provide sufficient resolution to address the phylogeny of the Delphacini. As expected from comparable analyses, non protein-encoding ribosomal DNA datasets provide most informational support for the deeper nodes of the phylogeny, while the nuclear protein-encoding genes provide greater resolution for the more recent nodes. Both kinds of molecular data show support for the morphological tree. The resultant combined consensus topology shows a series of intuitive clades, some of which imply ecological or host-based causative evolutionary mechanisms.

Structure of ovaries in apterygotous hexapods & its implications for the phylogeny of arthropods

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Evolutionary relationships among extant arthropods (insects, crustaceans, myriapods, and chelicerates) are not consistently resolved and remain controversial. Traditionally, it was believed that the largest arthropod subtaxon, Hexapoda (insects and their apterygotous allies, *e.g.* springtails and diplurans) is related to myriapods. Recent molecular data have challenged this alliance in favor of a close relationship between insects and crustaceans. In this context, we have decided to reinvestigate the structure of ovaries and process of oogenesis in representatives of wingless hexapods (springtails, diplurans and proturans), lower crustaceans (branchiopods) and myriapods (symphylans). Special attention has been focused on the following characters: organization of germ-cell clusters, presence (or absence) of mesodermal follicular cells, mode of yolk formation, synthetic activity of the oocyte, presence (or absence) of a mitochondrial cloud (*i.e.* Balbiani body) and origin of egg envelopes. Obtained results support close affinity between Hexapoda and Crustacea. Moreover, we found that myriapods and chelicerates (spiders and pantopods) share important "ovarian" synapomorphies.

Evolution of extreme prey specialization in bolas spiders

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Moth hunting bolas spiders construct some of the most specialized webs found within the Araneidae. These highly reduced webs are hypothesized to have evolved through a gradual reduction of the orb web within the Cyrtarachninae, which accompanied an increased specialization upon male moths as prey. However, this classic evolutionary story has never been empirically tested. We used both mitochondrial and nuclear DNA sequence data to construct a phylogeny of the Araneidae to test the monophyly of these moth specialists and to examine the pattern of evolution of bolas webs. We also compare our results with the current phylogenetic hypothesis of araneid relationships based upon morphology and behavior.

On the chemoecology of Danainae

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Danaine butterflies have multiple relationships to plants and utilize different secondary chemicals: larval hostplants which may contain cardiac glycosides (CGs), adult nectar plants, and plants providing pyrrolizidine alkaloids (PAs) which are taken up pharmacophagously by males. Some danaines make up neat biological stories, opening one's eyes for several phenomena and giving general lessons, e.g. on individual variation of traits. However, overviewing the entire subfamily in some detail unmasks our knowledge to be quite fragmentary and shows the common view of danaines to be full of incorrect paradigms and of speculations. CGs and PAs are both protective – but likely against different types of predators. Are CGs or PAs the basis for Batesian and Mullerian mimicry associations? Where lies the origin of the relationships to PA-plants? What drives elaborate sexual communication, what speciation? Open questions are many and multifold. Comparing the chemoecology of danaines (and combining morphological, ecological and chemical data) with those of arctiine moths reveals interesting parallels and similarities and challenges evolutionary hypotheses.

Molecular phylogeny of the Pieridae (Lepidoptera: Papilionoidea): systematics, higher classification & host plant associations

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Characters from the nuclear gene *Elongation Factor*-1 *alpha* were used to reconstruct the phylogeny of the butterfly family Pieridae. The sample included 86 species representing all but five currently recognised genera. 364 of the 1066 sequenced bp (34%) were parsimony informative, with most variable sites in the third codon position. The strict consensus of nine equally MP trees recovered four clades, plus two smaller, less well supported groups, and these show concordance with a higher-level classification based primarily on morphological characters. We use these data and others from the literature to present a revised classification of the Pieridae in which the Pierinae is divided into three tribes (Anthocharini sens. strict., Pierini sens. strict., Aporiini) and two informal groups (*Eronia* group, *incertae sedis*). The topology of our cladogram suggests the following evolutionary relationships among the higher taxa: Coliadinae + (*Eronia* group + ((Pseudopontiinae + Dismorphiinae) + (Anthocharini + (Pierini + Aporiini)))). Our data show that host plant use is correlated with phylogenetic relationship, with Brassicales feeding evolving once, Fabales feeding evolving at least twice, and Santalales feeding evolving independently three times as a derived state. The ancestral food plant was most likely Fabales, and host shifts appear to facilitate adaptive radiation within the family.

Phylogeny & the Evolution of Wings in extant Phasmatodea (Insecta: Polyneoptera)

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The Phasmatodea (stick and leaf insects) have recently been subject to several phylogenetic analyses, based on both morphological and molecular data. While a sister group relationship of the Nearctic genus *Timema* to all remaining Phasmatodea, the Euphasmatodea, has appeared uncontroversial, the primary internal splits of the Euphasmatodea remain debatable (Bradler et al. 2003 Ent. Abh. 61: 132-133). The first molecular analysis based on 18S rDNA, 28S rDNA and histone 3 sequence data proposes that several wingless taxa form the basal lineages of extant phasmids (Whiting et al. 2003 Nature 421: 264-267). This well supported finding implies that the ancestral phasmid lacked wings, followed by a long diversification of wingless taxa and multiple derivations of wings during phasmid evolution. Consequently, wings must have been recovered in those clades that contain fully winged or brachypterous species. The controversial assumptions regarding the phylogenetic relationships among basal phasmids highlight the need for future research. New phylogenetic trees inferred from morpho-anatomical characters of the adult form and egg capsule, additional molecular markers, and a larger taxonomic sampling (~100 species) are presented.

A New Family of Chrysidoid Hymenoptera from New Jersey Cretaceous Amber

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Two conspecific specimens of aculeate wasps found in Late Cretaceous amber from New Jersey (USA) cannot unambiguously be assigned to any known family. Features of the wing venation, mesosoma and mandibles indicate that they clearly belong in the Chrysidoidea. The wing venation is more complete than in any other member of the superfamily, but does not show the development of RS2 (as found in Plumariidae) sometimes considered to be primitive. The antennae have sparse long erect setae similar to those found elsewhere only in Plumariidae, and the pronotum is similar to that in Plumariidae and Scolebythidae. Cladistic analyses of the fossil in various combinations with the genera of Plumariidae, with groundplans for the families of Chrysidoidea and with the families of Aculeata, and using different character sets as appropriate, produced trees of similar length, with the fossil as the basalmost taxon in Chrysidoidea, the basal taxon within Plumariidae or as a taxon originating immediately after Plumariidae (and so sister to all other chrysidoids). The last topology (Plumariidae + (fossil + (Scolebythidae + (Bethylidae + Chrysididae) + (Sclerogibbidae + (Embolemidae + yinidae)))) was most often the most parsimonious and therefore preferred. It is thus proposed that the fossil taxon be recognised as a new family.

Molecular systematics of mimetic butterflies

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Since the time of Bates and Müller, the study of mimicry has advanced both theoretically and experimentally. Study of the taxonomy of butterflies was what led Bates to his discoveries in 1862, and as our work advances in this area, so do patterns of mimetic convergence become ever more complex. The molecular systematics of Heliconiina and Ithomiini will be reviewed. To a large extent, DNA evidence corroborates prior hypotheses based on morphology, but there are still controversies to be resolved and surprises yet to be revealed.

Moss bugs (Hemiptera: Peloridiidae): Systematics & biogeography

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Moss bugs or peloridiids constitute the only extant family of Coleorrhyncha, the sister group of Heteroptera or true bugs. The family has attracted a lot of attention due to its phylogenetic position within the Hemiptera, its Gondwana distribution as well as the scarcity of available specimens. Based on important collections the internal taxonomy and phylogeny was investigated. Seventeen genera are recognised containing almost 30 species. A cladistic analysis with using 28 adult morphological characters yielded 3 most parsimonious trees. The biogeographic analyses suggest that Peloridiidae diversified together with the break-up of Gondwana (New Zealand/New Caledonia, (South America+Australia). The position of New Caledonia is within and not sister to the New Zealand clade as is suggested by geology. Of particular interest is a taxon from Lord Howe Island which is closest related to a Northern Queensland genus.

Molecular evidence on phylogenetic relationships among major lineages of Pterygota

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Molecular data combined with morphology are leading to robust hypotheses for cladogenetic events among the basal pterygote lineages, and providing new insights into the diversification of insects. Multiple genetic markers from an extensive sampling of taxa were used to evaluate the monophyly of Paleoptera and to propose a phylogenetic hypothesis for the basal neopterous insect orders (="Paleoptera"), including the phylogenetic placement of Mantophasmatodea. Results from extensive phylogenetic analyses of the orders Ephemeroptera, Odonata, Plecoptera, Mantodea, Dermaptera, and Grylloblattodea will be presented.

Phylogenetic utility of mitochondrial genomes in resolving insect interordinal relationships

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The phylogenetic utility of the mitochondrial genome includes two main approaches – use of shared gene rearrangements as molecular synapomorphies and the use of sequence data in "traditional" phylogenetic analysis. In resolving the interordinal relationships of insects, gene rearrangements are virtually worthless; rearrangements whilst not uncommon do not unite any orders and are commonly variable within orders. The use of gene rearrangement data in resolving lower level relationships will be discussed. In contrast use of the mt genome sequence data in phylogenetic analysis shows considerable promise as a phylogenetic marker at the interordinal level. A phylogeny of insect orders using mitochondrial genome data will be presented.

Biodiversity, host plant associations & biogeography of the Heteroptera of continental Australia

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The true bugs of Australia are both diverse taxonomically and highly endemic. The true bug fauna is currently represented by 2000 species, within 64 families and almost 800 genera. In terms of percentage of true bug species richness globally, the Australian fauna is only 5% of the global estimate of 40,000 species. However, the Australian fauna is likely to be less than 50% described, with families such as the Miridae represented by at least another 1000 new species. In addition, the endemicity of the Australian fauna is extremely high, with 50% of the genera and 80% of the species found only in Australia. The diversity of true bugs is information exists regarding their host plant associations. These bugs have particularly close relationships with rosid and asterid angiosperms, suggesting a temporal correlation with the diversification of the angiosperms in the Cretaceous. The pentatomomorphans are often found in association with the "big two" of the Australian flora – *Eucalyptus* sensu lato and *Acacia* – and there is significant overlap in their distribution, both in terms of species richness and endemism. In addition, to these landscape patterns, the origins of the Australian true bug will be discussed in terms of historical biogeography.discussed in terms of host plant associations and biogeography. For the Pentatomomorpha, considerable

Phylogenetic position of the Hymenoptera within the Holometabola using entire mitochondrial genomes

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We sequenced the mitochondrial genome of the sawfly *Perga condei* (Insecta: Hymenoptera: Symphyta: Pergidae) and tested different models of phylogenetic reconstruction in order to resolve the position of the Hymenoptera within the Holometabola, using entire mitochondrial genomes. The mitochondrial genome of *Perga condei* had less compositional bias and slower rates of molecular evolution than the honeybee, as well as a less rearranged genome organization. Phylogenetic analyses showed that, when using mitochondrial genomes, both adequate taxon sampling and more realistic models of analysis are necessary to resolve relationships among insect orders. Both parsimony and Bayesian analyses performed better when nucleotide instead of amino acid sequences were used. In particular, this study supports the placement of the Hymenoptera as sister group to the Mecopterida.

Towards an evolutionary history of tortoise and leaf-mining beetles: systematics, major events & diversity (Coleoptera: Chrysomelidae: Cassidinae *sensu lato*)

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Systematics research in Cassidinae *s. l.* has been focused on three fundamental questions: their monophyly, position within Chrysomelidae, and internal relations. Borowiec (1995), Hsiao and Windsor (1999) and Chaboo (in press) addressed these issues. Their results are briefly compared. These studies help narrow the range of phylogenetic hypotheses and provide a historical context for evaluating evolutionary patterns in Cassidinae *s. l.* Evolutionary models using character mapping are developed to explain general trends and major innovations in cassidine diversification. Alteration of the adult head from prognathous to hypognathous appears in the early stages of their evolution. Compaction of the body, legs and elytral locking mechanisms are recurrent themes in adult morphology. Cassidine ecological guilds represent changes in life cycles, mouth morphology, and specialization on particular groups of host plants and specific parts of hosts (stem, leaf, or flowers). Fecal shields represent a crucial landmark in cassidine history. They are correlated with morphological innovations in larvae (urogomphi and anus) and pupae (urogomphi) and their appearance coincides with the transition from leaf-mining to open exophagy. A major radiation of ~3000 species followed the origin of shields.

Revision of the genus *Bambusiphaga* (Homoptera: Delphacidae) from Oriental Region, with descriptions of two new species

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The oriental planthopper genus *Bambusiphaga* Huang and Ding, 1979, feeding exclusively on Bambusoideae, is revised. Eighteen species were recorded formerly in the world, distributed mainly in China (16 species), Singapore (2 species), Malaysia (2 species), Philippines (1 species) and Northeastern Himalaya (1 species). In the present paper, 2 new species are described and illustrated in detail: *B. maolanaensis* Chen *et* Liang, sp. nov. and *B. pianmaensis* Chen *et* Liang, sp. nov.. Based on the characters of male genitalia, 20 species are divided into 7 groups of species. Two new synonymies are reported: *Malaxa herioca* Yang, 1989 is a junior synonym of *Bambusiphaga facia* Huang *et* Tian, 1980 and *Bambusiphaga latispina* Qin *et* Yuan, 1999 is junior synonym of *B. bakeri* (Muir), 1919. One new combination is proposed: *bakeri* Muir, 1919 is transferred from *Malaxa* Melichar to *Bambusiphaga* Huang *et* Ding. A key to the 20 species of *Bambusiphaga* is provided.

Biodiversity & ecology of marine water-striders (Gerromorpha)

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At least five families of Gerromorpha are found in various marine environments: -

1. Gerridae, with ca.700 described species, comprise more than 80 species (number in brackets) in 9 genera and 3 sub-families: Halobatinae – *Asclepios* (3), *Halobates* (46); Trepobatinae – *Pseudohalobates* (1), *Rheumatometroides* (8), *Stenobates* (10), *Stenobatopsis* (1), *Telmatometroides* (1), *Thetibates* (2); Rhagadotarsinae – *Rheumatobates* (11)
2. Hebridae, a small family, has only 2 *Hebrus* species found in mangrove areas in Singapore.
3. Hermatobatidae, an exclusively marine monotypic family, has 8 or 9 known *Hermatobates* species inhabiting coral rubble or rocky intertidal shores.
4. Mesoveliidae, another small family, is represented by four genera, *Darwinivelia* (3), *Mesovelia* (4), *Nereivelia* (1) and *Speovelia* (2). Little is known about their ecology.
5. Veliidae, with ca. 880 described species, comprise at least 81 species in 9 genera and 4 subfamilies: Haloveliinae – *Halovelia* (33), *Haloveloides* (13), *Xenobates*(20); Microveliinae – *Husseyella* (3), *Microvelia* (4), *Xiphoveloidea* (1), *Mangrovelia* (1); Rhagoveliinae – *Trochopus* (5); Veliinae – *Paravelia* (1).The Australasian region has an especially rich diversity of marine water-striders. The ecology of representative species will be discussed.

Creophilus (Coleoptera: Staphylinidae): phylogeny & historical biogeography

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The genus *Creophilus* Leach (Coleoptera: Staphylinidae: Staphylininae) contains 12 species worldwide, divided into the *C. maxillosus* (5 species), and *C. erythrocephalus* (7 species) species groups. This division also characterizes a biogeographical split within the genus corresponding approximately to Wallace's line: The *C. maxillosus group* is widespread throughout Europe, Asia, North and South America, and Madagascar while the *C. erythrocephalus* group is entirely austral. However the two groups overlap in Indonesia. Identification of synonymy and investigation of potentially new species from oceanic islands has helped stabilize the taxonomy. With a sound taxonomic framework a phylogeny of *Creophilus* is presented to determine if the two groups are monophyletic, and to assess the biogeographical implications of the phylogeny. More than 40 characters have been discovered that support the monophyly of *Creophilus* and the two species groups, both of which also share distribution patterns exhibited by many disparate groups. The combined phylogenetic and biogeographic analysis will determine which group is basal within *Creophilus* and hence determine the biogeographic origins of the austral species (ancestral versus derived), and to what extent vicariance or dispersal are sufficient to explain observed distribution patterns. The biogeography of the New Zealand species is emphasized.

Bat-moth interactions: tiger moth calls & acoustic mimicry?

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The interactions of insectivorous bats and their insect prey can be thought of as an evolutionary arms race. Bats evolved sophisticated high-frequency sonar and insects countered with the evolution of sonar detection devices – ears – and the ability to answer the echolocation cries with cries of their own. But why do moths answer? Are they producing sounds to startle bats, to jam their sonar, or are they warning the bats that they are unpalatable? Using a learning approach we have been able to query naïve big brown bats (*Eptesicus fuscus*) about clicking tiger moths. Our results indicated that many arctiids are indeed warning the bats of their unpalatability. In our studies the sounds produced by tiger moths are only deterrent when combined with unpalatability. Using the same learning methods we are now poised to test whether the sounds produced by tiger moths form an acoustic mimicry ring that may include other members of the Lepidoptera and sound-producing insects of several orders.

Sperm structure of Mantophasmatodea

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According to morphological characters, Mantophasmatodea are similar to the two better known stick-insects and Mantodea taxa. Sperm ultrastructure has indicated that the group is related to Mantodea, with which it shares, among other features, the presence of three connecting bands in the sperm flagellum. On the contrary, sperm structure does not support a relationship with Phasmatodea, a group with several autapomorphies. The study of spermatozoa of different species (*Karoophasma biedouwensis* and *Namaquaphasma ookiepensis*) from other regions of South Africa, kindly sent by Mike Picker, confirms the general structures described for *Mantophasma zephyra*; it allows also to identify new features not present in the previous species dealing with the peculiar chromatin condensation. Moreover, as in the previous species examined, it was established that spermatoozoa in the genital ducts and seminal vesicles are clustered in conspicuous spermatodesms consisting of hundreds of sperm cells. In these sperm bundles, the ordered sperm heads are joined by *zonulae adhaerentes*; these junctions have been established by the presence of actin filaments revealed by the positive reaction with the confocal laser-microscope, after phalloidin-rhodamine treatment.

Mantophasmatodea: genitalic morphology, species diversity, and intra-ordinal phylogenetic relationships

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Presently 13 extant (all African) and 1 fossil species (European) of Mantophasmatodea are known: South African *Austrophasma* (3 species), *Lobophasma* (1 species), *Hemilobophasma* (1 species), *Karoophasma* (2 species), and *Namaquaphasma* (1 species); Namibian *Sclerophasma*, *Mantophasma*, *Praedatophasma*, and *Tyrannophasma* (1 species each); Tanzanian *Tanzaniophasma* (1 species); Baltic amber *Raptophasma* (1 species). Species distinction is based on structure/shape of clasping cerci, vomeroid (sclerotized projection above genitalia), and genitalic sclerites and projections in males, shape of subgenital plate and spermathecal bulb in females, and mottled versus striped colouration of eyes. Body colouration varies strongly in most species. Cladistic analyses using 16SrDNA and COI (*Mantophasma*, *Praedatophasma*, *Tanzaniophasma* not included) combined with tentative placement of *Mantophasma*, *Tanzaniophasma*, and *Namaquaphasma* based on genitalic morphology results in [*Tanzaniophasma* + (*Tyrannophasma* + ((*Sclerophasma* + *Mantophasma*) + (*Austrophasma* + *Lobophasma* + *Hemilobophasma* + *Karoophasma* + *Namaquaphasma*))). Further yet undescribed species have been collected in South Africa in 2003, and are included in the phylogenetic analysis. Relationships among South African genera have remained insufficiently resolved. At least the South African species are highly endemic, rarely co-occurring in the same locality.

Phylogeny of the true water bugs (Nepomorpha) based on 16S and 28S rDNA & morphology

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Morphological characters and molecular sequence data are for the first time analysed separately and combined for the true water bugs (Hemiptera-Heteroptera, infraorder Nepomorpha). Data from 40 species representing all families are included together with 2 outgroup species representing the infraorders Gerromorpha and Leptopodomorpha. The morphological data matrix consists of 65 characters obtained from literature sources. Molecular data include approximately 960 bp from the mitochondrial gene 16S and the nuclear gene 28S rDNA. The morphological dataset was analysed using maximum parsimony and the combined morphological and molecular dataset was analysed using direct optimisation (POY). The preferred phylogeny supports the monophyly of all currently recognised families of Nepomorpha, and of the superfamilies Nepoidea (Nepidae + Belostomatidae), Corixoidea (Corixidae), Ochteroidea (Ochteridae + Gelastocoridae), Notonectoidea (Notonectidae), and Pleoidea (Pleidae + Helotrephidae), but not the monophyly of the Naucoroidea (Naucoridae + Aphelocheiridae + Potamocoridae). Our hypothesis concur with Mahner (1993) in the basal placement of the Corixidae as sister group to the remaining nepomorphan superfamilies except the Nepoidea, but differ in the placement of the Ochteroidea as sister group to the Notonectoidea + Pleoidea. Finally, our analysis strongly supports a sister group relationship between the families Aphelocheiridae and Potamocoridae.

Biology & diversity of the plant-mining family Opostegidae

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Although essentially cosmopolitan in distribution, the monotrysian family Opostegidae has been poorly surveyed and studied largely due to inherent difficulties in collecting both larvae and adults. Recent studies involving mostly the Oriental and Neotropical Regions over the past five years have nearly doubled the known species. Currently 7 genera and 193 species are recognized worldwide. Monophyly of the Nepticuloidea (Nepticulidae + Opostegidae) is well supported by 9 morphological synapomorphies. The Opostegidae comprise an isolated derivative of Nepticulidae as indicated by more than 18 larval and adult synapomorphies. Only 8 species in 4 opostegid genera have been reared, half of these within the endemic Hawaiian leaf-mining genus *Paralopostega*. Plant hosts of 5 additional species have been tentatively associated. All records demonstrate a broad range of host utilization involving possibly 8 plant families dispersed among 7 dicotyledonous orders. The univoltine life cycle for Opostegidae typically consists of: a) egg deposited on host leaf, b) a brief initial linear leaf-mining phase leading to c) an extended period of stem-mining followed eventually by d) pupation in a silken cocoon in the soil. Restricted leaf-mining, as observed in *Paralopostega*, represents a derivation from the norm.

On the origin of worldwide distributions in butterflies

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The emphasis laid on vicariance to explain disjunct distributions during the last 50 years, originating from an inductive way of reasoning, has led to unlikely scenarios including derivation from Pangea as a general explanation of (semi-)worldwide distributions. A deductive way of reasoning, testing alternative scenarios, seems a more productive approach. Only scenarios differing in time frame can be tested against each other. Because of the paucity of fossil butterflies the estimation of divergence times must be derived from the application of a molecular clock. Calibration of the molecular clock is crucial. Obviously if we want to test a vicariance scenario, the age of the geological vicariance event cannot be used to calibrate the clock. If a fossil can be used, it gives an indication of the minimum age of the taxon under study. Better calibration points, but obviously only applicable in particular instances are the age of oceanic islands and the age of foodplants. In both cases we get an estimate of the maximum age for the divergence (speciation event). Examples are given from Hesperiidae, Papilionidae, Lycaenidae and Nymphalidae. The impact on understanding early butterfly diversification is mentioned.

The phylogenetic analysis of complex mitochondrial gene rearrangement characters

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The discovery of complex gene rearrangements raises analytical issues. In particular, we and others have documented the situation wherein related taxa have one of a number of gene rearrangement variants. These have been variously and inconsistently interpreted as independent rearrangements, as a single, general rearrangement, or as a series of intermediates. No empirical data have yet been directed at assessing which of these approaches most closely mirrors biological reality. We assessed this issue amongst a group of braconid wasps at the cytochrome oxidase II/ATPase 8 gene junction, which ancestrally contains the tRNA genes for lysine and aspartate. Initial screening of this mitochondrial region revealed two variants of what appeared to be a compound rearrangement – some braconid wasps had the aspartate tRNA gene inverted, others had both the aspartate tRNA gene inverted and the histidine tRNA gene translocated to this gene junction. An independent analysis of phylogeny (16S, 28S, morphology) resolved each variant as belonging to discrete monophyletic groups, along with other braconids that retain the ancestral genome organization. This study indicates that the only analytical approach that would have resolved phylogeny consistent with the independent data is to treat these two variants as separate unordered characters.

Phylogeny of Chrysomelidae based on two molecular markers & morphological characters

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The Chrysomelidae are one of the largest families of beetles and although there are many published hypotheses for the relationships among the subfamilies, their relationships continue to be controversial. A phylogeny of Chrysomelidae using representative taxa from all subfamilies and based on 18S and 28S-D2 molecular markers and morphology is presented and discussed. Megalopodidae and Orsodacninidae are studied as outgroups. Phylogenetic analyses are performed using parsimony and maximum likelihood techniques. Support for the monophyly of well-recognized taxonomic groups is discussed and evaluated in light of recent literature. Phylogenies resulting from separate and combined analyses of each data set are presented; the 18S data set is not ideal for resolving the relationships among the subfamilies of Chrysomelidae. The hypothesis that the chrysomelid radiation was too rapid and too ancient to be effectively recovered using exclusively molecular data is explored. Support for differing hypotheses explaining the evolution of morphological characters such as, bifid setae, the kotopresse, and Maulik's organ (metafemoral apodeme) are explored. A reduced taxon data set involving a third molecular marker (cytochrome oxidase one) may be presented. This is the first analysis of the Chrysomelidae to use more than one molecular marker.

Phylogeny of the flea beetles (Chrysomelidae: Galerucinae: Alticini) based on three molecular markers

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The flea beetles (Galerucinae: Alticini) are notable in the lack of taxonomic consensus describing the relationships between the genera and their relationship to the traditional "galerucines". As the first part of a larger study of the phylogeny of the flea beetles, a phylogeny of the flea beetles using over 100 representative genera from all continents and based on fragments of large and small subunit nuclear ribosomal DNA (18S V7-9 and 28S-D2) and cytochrome oxidase I molecular markers is presented and discussed. Genera from the Galerucinae s.s. and 9 Chrysomelinae genera are also analyzed. Phylogenetic analyses are performed using parsimony and maximum likelihood techniques; the alignment was generated using secondary structural models. Support for the monophyly of the well-recognized taxonomic groups, Oedionychina, Monoplatina and Disonychina is discussed. Support for other groupings previously, recognized or not, is also discussed. Phylogenies resulting from separate and combined analyses of each data set will be presented; the 28S data set offers significant synapomorphies for some clades. Hypotheses describing the loss of Maulik's organ (the metafemoral apodeme) and evolution of endophagy in the Galerucinae s.l. will be presented and discussed.

The challenge of insect taxonomy in developing countries: A case study of the genus *Oribius* Marshall (Coleoptera: Curculionidae) in Papua New Guinea

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Taxonomists of the developing tropical nations have a constant challenge of how to identify their diverse native faunas, without having to rely entirely on international colleagues. A study of *Oribius* Marshall (Curculionidae: Celeuthetini), a genus with agricultural importance endemic to New Guinea and Australia, is used as a case study to illustrate this issue. Objectives of the project were to provide working keys to pest species of *Oribius* for agricultural researchers, while at the same time developing a taxonomic synopsis of the remainder of the genus. The study was conducted in Papua New Guinea by National scientists, with collaborative support from Australia. Operational and taxonomic challenges included the limited availability of: national systematists, keys, type specimens, literature and operating funds. Ultimately the identifications of five out of seven pest species were confirmed: the remaining two pest species may be undescribed. Very limited advances could be made on the remaining 40+ non-pest species. This project demonstrates that development of local expertise is a better model than the historical one of simply sending specimens off for identification to international experts. However, it does highlight the enormous need for true collaboration between scientists from developing and developed countries.

Evolutionary trends in derelomine flower weevils: from associations to homology

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Derelomine flower weevils (Curculionidae: Derelomini) have longstanding, remarkably specialized associations with palms and other plants such as members of the Neotropical family Cyclanthaceae. Recent morphological phylogenetic analyses, particularly of taxa related to *Phyllotrox* Schoenherr and *Celetes* Schoenherr, are starting to provide a context within which these life histories can be analyzed and explained. The results send a mixed, though exciting message. Host plant associations - understood at an appropriately deep level - are often as informative phylogenetically as other traits. Yet the picture emerging from natural history studies is more complex. Certain lineages of weevils are not only restricted to a limited set of related host plants. They also display an array of morphological, ecological, and behavioral adaptations that are *correlated* with phylogenetic transformations in inflorescence morphology and phenology. A truly adequate language for these phenomena requires that we regard "host plant associations" merely as a starting point for evolutionary analyses. More in-depth studies of selected interactions should aim at making narrower homology statements, and view the plants as entities that have come up with their own responses to a range of beneficial or detrimental interactors. In the case of derelomine flower weevils, this approach will probably teach us more.

A phylogenomic approach to spider silk gene diversification

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Spiders are unique in their ability to spin multiple types of task-specific silks. Each type of spider silk is composed of exceptionally long protein molecules (fibroins) that are synthesized in specialized silk glands. Several lines of evidence demonstrate that a large gene family encodes the different fibroins. To date, only a small fraction of the silk gene family has been characterized at the nucleotide level. Moreover, the silk genes sequenced thus far are sampled from a small number of species. In order to reconstruct the evolutionary history of the spider silk gene family, we are using a comparative approach to isolate novel silk gene sequences from a phylogenetically broad spectrum of spiders. Specifically, we have constructed and are screening cDNA libraries for novel silk transcripts from diverse taxa. We have also sequenced ESTs from these cDNA libraries in order to develop nuclear markers from which we are generating a spider species-tree. Using the species-tree in conjunction with the silk-gene tree, we will infer the number of duplications and losses of silk gene homologs.

Mitochondrial phylogenetics of UK Eurytomids

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The aim of this study was to investigate the taxonomic limits of species within four Eurytomid genera namely *Eurytoma* Illiger, *Tetramesa* Walker, *Ahtola* Claridge and *Sycophila* (Walker). In order to further clarify the taxonomic status of the genera: including *Tetramesa*, *Eurytoma*, *Ahtola* and *Sycophila*, mitochondrial DNA sequence analysis revealed differentiation between the above Eurytomidae genera. With the sequence data, the monophyletic status of three of these genera is well supported by all methods of phylogenetic reconstruction. Only *Tetramesa*, which is by far the most specious taxon studied here, seems polyphyletic, but even in this case, there are certain relationships within the group that are interesting from both a morphological and host preference point of view. For example, *T. petiolata* (Walker) and *T. airae* (von Schlechtendal), which are both herbivores of *Deschampsia cespitosa* (L.) (Beauv.) group together and two clusters contained all species that use *E. repens*, i.e. *T. linearis, T. hyalipenis* (Walker) and *T. cornuta* (Walker). Another result of this analysis supports the validity of *Ahtola* as a separate genus which was previously uncertain.

Molecular phylogeny of Micropterigidae (Lepidoptera)

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Micropterigidae are tiny, sedentary, forest moths consisting of about 120 species in 10 genera. The family has been provisionally divided into two groups, the '*Micropterix*-group' (*Micropterix* only) and the '*Sabatinca*-group' (nine other genera), but their relationships remain obscure. Our study sequenced portions of the mitochondrial 16S rRNA gene of about 50 species in 12 genera including three unnamed generic taxa. Phylogenetic analyses revealed five major clades in this family. The analysis confirms the monophyly of *Micropterix* (100% support) in the Northern Hemisphere. However, the '*Sabatinca*-group' taxa comprise a complex of 4 clades of which the majority are Southern Hemisphere, with the exception of Asian genera (*Neomicropteryx*, *Palaeomicroides*, *Paramartyria*, and *Vietomartyria*) and North American *Epimartyria,* which formed a monophyletic group (bootstrap value 100%). The remaining southern taxa form 3 clades: *Sabatinca s. str*. (100% support) restricted to New Zealand and New Caledonia, a weakly connected group (53% support) containing "*Sabatinca" porphyrodes*, the S. American *Hypomartyria*, the S. African *Agrionympha*, two undescribed species from Madagascar and two Costa Rican unnamed genera, and finally a distinctive Australasian group with species in Eastern Australia, New Zealand and New Caledonia (100% support). Although the basal relationships among these clades remain unclear, it is suggested that divergence of some major lineages in Southern Hemisphere can be related to the breakup of Gondwana.

A preliminary phylogeny for the subfamily Chrysomelinae (Coleoptera: Chrysomelidae)

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The morphological characters of insect immatures are often used in systematic analyses of insect relationships, but rarely given parity with adult morphological characters. One reason for this inequality is that immatures are often unknown; another is that adult characters are considered to carry greater phylogenetic weight. In Coleoptera, larvae provide considerable information in phylogenetic analyses of the Adephaga, but are little used elsewhere. The Chrysomelidae, subfamily Chrysomelinae, is a large group of phytophagous beetles with a classification that has changed little over the past 150 years. The known larvae are external plant feeders and easy to rear. Many species have been reared and many authors, including W. Hennig, have noted that larval morphology does not match the adult classification currently in use. Here we explore the use of larval morphology in chrysomelid systematics, using the Chrysomelinae as an exemplar taxon. The contrasting adult and larval morphological information is compared with phylogenetic signal provided by DNA sequence data.

Monophyly & phylogenetic relationships in basal Dermaptera

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The monophyly and phylogenetic relationships in Dermaptera are examined with new character systems and characters previously used for identification purposes. Dermapteran monophyly is difficult to assess because some basal taxa reduced character systems with compelling synapomorphies. Examples are highly modified hind wings, which are often reduced, and the ovipositor. The situation is further complicated through the Arixenina and Hemimerina, which live on bats (Sarawak) and hamster rats (sub-Saharan Africa). They are morphologically so strongly modified (wingless, cerci not forceps-shaped, viviparous) that at times they were ranked as separate ‚orders'. Recent research, however, indicates close relationships to Spongiphoridae and Apachyidae. Characters for monophyly, including Arixenina and Hemimerina, are lateral overlapping of abdominal tergites and sternites and holocentric chromosomes, both character states only occur in distantly related taxa. Many characters are not convincing as dermapteran autapomorphies as they also occur in potential sister groups of Dermaptera, while evidence from them is conflicting: single-annulus cerci (‚one segment'), maternal care, hind wing folding. For the first time the ovarian, spermathecal and tarsal structure of a number of basal taxa is examined and interpreted phylogenetically. These new character systems provide support for currently recognized ‚families' as monophyletic taxa.

On the fly: an interactive identification key to Australian fly families

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There are 104 families of flies known from Australia and the only comprehensive identification guide to these groups is found in *The Insects of Australia* (CSIRO). This key is technically challenging and difficult for non-taxonomists to use. We are currently developing a computer-based interactive (polyclave) key to Australia's fly families using Lucid©, called "On-The-Fly". The key divides the families into four broad groupings (Lower Diptera, Lower Brachycera (including Lower Cyclorrhapha), Calyptrata and Acalyptrata). The family keys are accessed via a key to the four groupings, and a section to assist in distinguishing true flies from similar insects belonging to other orders. On-The-Fly is richly illustrated with images of character states, and live images of representative species. There will be a taxon information page on each family, including information on diversity, biology, ecology and pest status. The key is a complete educational tool with general information about fly biology, classification, collection, curation, links to literature and websites. The software package is greatly enhanced for education with the inclusion of an interactive glossary of fly anatomy for use in conjunction with the key. On-The-Fly is under development and a beta version will be demonstrated.

Comparison of life history traits including diapause regulation among four species of water striders, *Aquarius paludum*, *Gerris latiabdominis*, *G. nepalensis* & *G. gracilicornis* living at 33ºN

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The four species of Japanese common water striders, *Aquarius paludum, Gerris latiabdominis, G. nepalensis and G. gracilicornis* overwintered as adults which entered reproductive diapause. Even though populations of the four species were living in Kochi (33ºN) located in a critical area between warm temperate and subtropical zones, they showed very high variation in the duration of diapause period, from three (*G. nepalensis*) to nine or ten (*G. gracilicornis*) months. All of *G. gracilicornis* and majority of *G. latiabodominis* adopted obligatory diapause, although part of individuals in *G. latiabdominis* populations showed facultative diapause: long-days induce reproduction and short-days diapause. *Aquarius paludum* and *G. nepalensis* entered reproductive diapause, when they were grown under short days. *Gerris nepalensis* and *A. paludum* adopted facultative diapause, responding to photoperiod and temperature to make up bi- or trivoltinism in Kochi. Eggs and adults of *A. paludum* inhabiting temporary to permanent water surfaces had higher drought-hardiness than those of *G. nepalensis* which lived exclusively permanent habitat. The short active period shown by *G. latiabdominis* seems to be caused by short season of March to July when paddy fields as their main habitat filled with water. The four species showed a high variety of life history traits including diapause regulation.

A cortical conundrum: New Zealand's scribbler & the biogeography of Cecidosidae (Lepidoptera: Incurvarioidea)

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The long bark-mines made by New Zealand's 'kamahi scribbler' on the trunks of several species of rainforest tree were first noticed in the 1950's, but the insect responsible was not reared through until 1990. It proved to be a moth belonging to the primitive (non-ditrysian) superfamily Incurvarioidea, and remains the only known member of this superfamily native to New Zealand. Study of larval, pupal and adult morphology revealed that it shared several synapomorphies with the Cecidosidae, a family otherwise only known from South Africa and South America, where the larvae form galls on Anacardiaceae. The New Zealand 'scribbler' (*Xanadoses nielseni* Hoare & Dugdale) has recently been described as a cecidosid, and in a tentative phylogeny is considered to represent the basal taxon in the family. Whilst the proposed phylogeny is congruent with the timing of the break-up of Gondwana, the apparent absence of Cecidosidae from the rich incurvarioid fauna of Australia remains to be explained.

Phylogeny & biogeography of Archostemata (Coleoptera), with special emphasis on the Australian monophyla

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With 40 extant species the Archostemata are the smallest suborder of Coleoptera. Nevertheless, these species are important for the study of the evolution of Coleoptera because they seem to have hardly changed morphologically during the last 200 Mio. years. The oldest known fossil beetles in many respects are very similar to the extant Archostemata. By studying this taxon we can expect to significantly increase our knowledge about the original morphology of Coleoptera. Another aim of this investigation was to reconstruct the species level phylogeny of the Archostemata. For this purpose, an investigation of their morphology was done, including high-resolution X-ray tomographic studies of a number of species. This method was used to gain as much information as possible from the few available specimens of these rare species. A main result is the identification of an Australian monophylum. This evolutionary young taxon includes *Distocupes*, *Ascioplaga* and *Adinolepis*, which are endemic for Australia and New Caledonia. Their most closely related group are the *Tenomerga*-species from Asia and South-East Asia. The evolutionary history of these groups goes back to the late Cretaceous. Nevertheless, at least some of the extant species might have had their origin at the end of the last ice

Successes & snags of continental island arthropod inventories: case studies in New Caledonia, Madagascar, & Fiji

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Continental island bioinventories can provide a wealth of information about the evolution of entire groups of poor dispersing organisms that arose before or during the Jurassic by relating evolutionary radiation and extinction events to the separation and attachment of drifting continents from or to other land masses. Inventorying arthropods has been an ongoing activity of ours. Over the past 15 years, much of our efforts have focused on flies (Diptera) of islands confirmed or believed to be continental in origin. We have conducted bioinventories on three island chains, New Caledonia in the 1990s, Madagascar in the late 1990s (still ongoing), and Fiji in 2003 (just getting underway). As bioinventories have transitioned from one island to another, we have gained considerable knowledge of how to conduct them and how to increase the probabilities of collecting not just greater quantities of material, but encountering those species that seem rare. We will share this knowledge and inform the audience of the difficulties in properly coordinating specimen storage, sorting, curation, and identification tasks. Beyond that we provide evidence of the importance of these inventories to global concepts in the evolution and biogeography of specific groups of flies.

Preliminary investigations on the evolution of true land bugs (Hemiptera: Heteroptera)

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The reproductive morphology, embryogenesis and characteristic of eggs in two infraorders of the hemipteran suborder Heteroptera were studied in representatives of five families in Cimicomorpha and 11 families of Pentamomorpha. All together, 65 species from 45 genera belonging to the families Miridae, Nabidae, Phymatidae, Reduviidae, Tingidae, (Cimicomorpha); as well as Acanthosomatidae, Alydidae, Aradidae, Pyrrhocoridae, Rhopalidae, Scutelleridae, and Thyrecoridae (Pentamomorpha) were investigated. The bugs were collected in the Nearctic and Palearctic regions, mainly in Canada, England and Iran. A comparative study of male and female reproductive systems, male aedeagus and female ovipositor, development of eggs, egg burster, and characteristics of egg shells have been studied. The results indicated the two infraorders are probably sister groups, and various shaped eggs in these groups have evolved independently, and along different path ways from an ovoid ancestral egg. The collected data is a valuable resource for tracing the evolution and phylogeny of the above two heteropteran infraorders.

Australasian Aulacidae (Hymenoptera: Evanioidea), endoparasitoids of wood-boring beetles & wasps

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The Aulacidae is a family of parasitic wasps that are endoparasitoids of wood-boring sawflies (Xiphyiidae) and beetles (Cerambycidae and Buprestidae). Until recently, the Australian fauna comprised 34 described species (18 *Aulacus* and 16 *Pristaulacus*), primarily from the wetter forested areas of the east coast of Australia. Just one species from New Guinea was described and they were unknown from New Caledonia, New Zealand and the other island groups of the south-west Pacific. However, based on specimens in museum and other collections, the aulacid fauna of the region is likely to exceed 100 species. The distribution of the group has been substantially expanded with new species known from south-west Western Australia, the arid interior of Australia, and from New Caledonia. This paper presents data on the distribution of these wasps in the Australasian region and discusses a range of characters and character states to be used in a phylogenetic analysis. Characters investigated include those associated with sculpturing, wing venation, and the ovipositor guide.

Phylogeny of the Gelechioidea: evolution of larval food substrate use

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The phylogeny of the lepidopteran superfamily Gelechioidea was estimated using 193 characters derived from larval, pupal and adult morphology and larval ecology. 143 species represented the diversity of the putative Gelechioidea in the analysis, and further 13 species representing three monotrysian and eight other ditrysian families were included to test the monophyly of the Gelechioidea. The Gelechioidea was nested within Apoditrysia instead of being its sister group as previously suggested. The well-resolved cladogram suggested a largely different concept of interrelationships of constituent gelechioid families as compared to other studies. The Gelechioidea was basally divided into two main lineages, herbivory as the ancestral larval feeding strategy of one, and, depending on character optimization, either saprophagy or herbivory of the other. Therefore it is not possible to unambiguously determine which of these traits is the ancestral one in the Gelechioidea. It is also sometimes questionable whether such a classification of the food substrate can be applied, .e.g. if the larva uses dry seeds and other dry vegetable matter. Detritivory/fungivory may have evolved only twice within Gelechioidea, though the evolution of larval food substrate use has been quite dynamic with frequent reversals.

Basal phylogenetic relationships in Dictyoptera

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Monophyly of Dictyoptera is supported through (1) perforation in tentorium; (2) female subgenital plate by coxosternum 7, bearing a dorsal vestibular sclerite; (3) packing of eggs in an ootheca built by accessory glands of abdomen 9; (4) paratergal insertion of abdominal spiracle dilators. For reconstructing dictyopteran phylogeny male and female genitalia (mostly reduced in Isoptera) and the proventricle are most informative. Cladistic analysis of morphological and life-history characters strongly supports Isoptera as the sister group of Cryptocercidae, hence as nested within Blattaria. Cladistic analyses using 107 characters of the exoskeleton and musculature of male genitalia suggest [Blattidae + (Tryonicidae + ((Cryptocercidae + (Lamproblattidae + Polyphagidae)) + (Anaplectidae + (paraphyletic Plectopterinae + (Blattellinae + Nyctiborinae + Ectobiinae + Blaberidae)))))] for Blattaria, and [Mantoididae + (Chaeteessidae + (Metallyticidae + remaining Mantodea))] for Mantodea; however, the taxon sample (currently 25 taxa) will have to be extended. Sequences from 12S, 16S, and 18SrDNA and COII also suggest Isoptera to be sister to Cryptocercidae, and sequences from *Blattabacterium* fat-body endosymbionts confirm this; however, the Cryptocercidae-Isoptera clade appears as sister to Blattidae.

Mantophasmatodea: important aspects of the skeleto-muscular system and internal organs

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Mantophasmatodea are Pterygota due to the absence of a non-cuticular endoskeleton and lateral parts of abdominal tergum 11, and Neoptera due to the presence of typical abdominal spiracle muscles and apodemes. Morphological peculiarities (autapomorphies) are: (1) specialized antenna structure: basiflagellomeres without constrictions between them, only with coarse setation, the distal ones showing a secondary subdivision; 7 distiflagellomeres, 1st very long, 2nd very short, all separated by constrictions and with fine setation; (2) strongly angled course of subgenal sulcus, depending on a dorsal shift of the origins of the anterior tentorial arms; (3) medioventral projection on male subgenital plate (for umming); (4) conspicuously blunt tip of gonapophyses 8 in ovipositor; (5) small dorsal projection in membrane beyond 3rd tarsomere. Proventricle resembling that in Notoptera. Circulatory organs: no lateral arteries in abdomen; antennal circulatory organs consisting of non-pulsatile ampullae and vessels with a characteristic double-layered wall (inner epithelium strongly resembling the transport epithelium so far reported only from antennal vessels of Dictyoptera); female cerci without distinct circulatory organ.

Early embryonic development of basal Lepidoptera

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Lepidoptera are generally regarded as the insect group whose embryogenesis is well known. From the phylogenetic standpoint, however, the materials have been concentrated on the highest group or Ditrysia, and no embryological work appeared on the lower non-ditrysian Lepidoptera until about 25 years ago when we started to wok on the embryogenesis of the basal Lepidoptera. Since then much information has been accumulated on the embryogenesis of such non-ditrysians as *Neomicropteryx* (Micropterigidae), *Eriocrania* (Eriocraniidae), *Endoclita* (Hepialidae), *Mnesarchaea* (Mnesarchaeidae), *Stigmella* (Nepticulidae), and *Nemophora* (Adelidae). Then large differences have been found especially in the early developmental processes among these and ditrysian Lepidoptera and these embryological data are also expected to afford significant means for tracing the phylogenetic relationships among major lepidopteran lineages. We will discuss some major differences and their phylogenetic implications: (1) The thickness of the blastoderm, very thin in *Neomicropteryx*, relatively thick in *Eriocrania*, *Stigmella*, and *Nemophora*, but very thick in *Endoclita* and *Mnesarchaea*. (2) The shape and size of the germ disk. (3) The mode of formation of the germ rudiment and embryonic membranes, the invaginate type in *Neomicropteryx*, *Endoclita*, and *Mnesarchaea*, the amnio-serosal fold type in *Eriocrania*, and the fault type in *Stigmella* and *Nemophora*.

Morphology of insect tagmata and phylogenetic implications

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The distinctive body division into three tagmata (head, three-segmented thorax, eleven-segmented abdomen) still represents the main argument for the monophyly of the Hexapoda. Recent molecular studies, however, challenged the view that the corresponding tagmatization of the trunk is an apomorphy. Presently competing hypotheses either assume that hexapods are derived from an already tagmatized ancestor or suggest paraphyly of Hexapoda. According to the latter, the hexapod tagmosis evolved convergently in at least Collembola and the "true insects" (i.e. Ectognatha). In order to test these hypotheses, the morphological evidence is revised. Special attention is awn on the skeleto-muscular system of primarily wingless insects, on which new data are presented. Comparisons with the state in Protura and Diplura permit to reconsider their relationship to the remaining hexapods. Insights obtained are discussed with respect to current assumptions that Remipedia or Malacostraca within the previous Crustacea may form the insects' closest relatives.

A phylogenetic study of the ant- and bee-loving soft scale insects of the world (Hemiptera: Coccoidea: Coccidae)

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In the Coccidae, the subfamily Myzolecaniinae is composed of soft scales that exhibit a strong bond with ants. These coccids are one of the most derived groups in the Coccidae, having morphological features that have evolved in such a way that these scale insects can no longer survive without attendance by their ant symbionts. The subfamily Myzolecaniinae (*sensu* Hodgson, 1994) contains the following genera: *Akermes*, *Alecanium*, *Alecanopsis*, *Cribrolecanium*, *Cryptostigma*, *Cyclolecanium*, *Halococcus*, *Houardia*, *Megasaissetia*, *Myzolecanium*, *Neolecanium*, *Paractenochiton*, *Pseudophilippia*, *Richardiella*, *Toumeyella* and *Xenolecanium*. The Myzolecaniinae have a worldwide distribution and are thought to be composed of several non-related genera which share common characters such as: the reduction in size of legs, antennae and anal tube; the increase in size of spiracles; lack of eyespots; and other shared characters that could be a result of parallel evolution. The objectives of the present study were: 1) to resolve classification conflicts within the subfamily Myzolecaniinae; and 2) to identify monophyletic groups and lineages within the subfamily. A total of 100 morphological characters from adult females and first instar nymphs were used. One new subfamily, the Toumeyellinae, and the *Alecanopsis*-group are proposed.

Some key events in the structural evolution of basal heteroneuran Lepidoptera

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While the phylogeny of the homoneurous-grade moth superfamilies has been at least preliminarily resolved, much uncertainty remains about the interrelationships of the non-ditrysian and basal ditrysian Heteroneura. For some notable character transformations plesiomorphic and apomorphic states are known to occur, respectively, among the homoneurans and the advanced ditrysians, and multiple discrete steps may be recognizable in the transformation series. Establishing the occurrence of the apomorphic state(s) in basal heteroneurans is, then, of considerable interest for phylogeny reconstruction. Cases discussed include (1) the proboscis base and its extrinsic musculature, which in basal glossatans differ but little from the generalized endopterygote type ascribed to the lepidopteran ground plan, but which are highly modified in 'typical' (ditrysian) Lepidoptera; (2) the ocellus with its diversity in 'vitreous body' and cornea morphology; (3) the testicular sperm with the complex 'lacinate appendages' in 'typical' ditrysians.

The basal splitting event recognizable in extant winged insects: the morphological evidence

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Morphological evidence bearing on the question of the relations between the conventionally recognized extant basal pterygote assemblages (mayflies, dragonflies and Neoptera) will be summarized; it has been awn from a suite of character sets in both immatures and adults. The bulk of the morphological evidence so far considered supports the monophyly of the taxon 'Metapterygota' (agonflies + Neoptera). Morphological evidence supporting the principal alternative hypothesis of a monophyletic 'Palaeoptera' (mayflies + dragonflies, retrieved in recent rDNA analyses) is more ambiguous, and that supporting a monophyletic 'Chiastomyaria' (mayflies + Neoptera) even more so. While neopteran monophyly has been accepted by most insect systematists, it has been questioned for some time by Rasnitsyn and his coworkers, and indeed it deserves more scrutiny.

The origin & early history of leaf mining

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Leaf mining is the only major insect functional feeding group whose earliest occurrence is documented in the Mesozoic, rather than the Paleozoic. Currently dominant on angiosperms, the earliest demonstrable fossil leaf mines are present on a variety of gymnospermous seed-plant clades, including the ginkgophytes *Paraginkgo* and *Sphenobaiera*, the probable bennettitalean *Dejerysea*, the herbaceous broad-leaved conifer *Heidiphyllum*, the ubiquitous peltasperm seed-fern *Dicroidium*, and a fern. These mines occur in the Late Triassic Molteno Formation in South Africa, dated at approximately 225 Ma and precede the earliest angiosperms by 90 million years. Culprits are probably coleopterans. With the exception of two other Gondwanan Triassic occurrences, a leaf-mining hiatus persists until the Jurassic-Cretaceous boundary, from which sinusoidal mines are found on another peltasperm seed-fern, *Pachypteris*, from the Battle Camp Formation of northern Queensland, Australia. The Dakota Formation of the United States, redated as latest Early Cretaceous, has yielded 10 leaf-mine types, representing the lepidopteran families Nepticulidae and Gracillariidae. They colonized basal dicotyledonous clades, including the Platanaceae, and taxa best assigned to the Chloranthaceae and Lauraceae. These data indicate that leaf mining first originated in Gondwana, opportunistically invading several gymnosperm and fern clades, and later underwent a radiation on basal dicotyledonous angiosperms in Laurasia.

Rewards & challenges of global, collaborative taxonomy: the paradigm shift offered by PEET

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The history of taxonomy is dominated by single author contributions, ranging in scope from single species descriptions to monographs. The NSF PEET program offers taxonomists the opportunity to explore a paradigm common in other sciences: multi-nodal, multi-authored collaborations. A major challenge for the 10-year therevid PEET project was coordinating global collaborative taxonomies of this medium-sized fly family (4,000 species). An international team including principal investigators, postdocs, 7 PhD and 4 Masters Degree students have monographed taxonomic "parcels", in a largely bottom-up approach. To date, over 47 papers on systematics, biology, biogeography, and morphology of therevids have been published through this global PEET collaboration, and a world catalog of Therevidae is planned. Tension developed between the necessity for students to work on manageable groups and the need for a family-level overview. The phylogenies for the family were developed through an iterative process, from both molecular and morphological datasets, with feedback between top-down and bottom-up approaches. Collaborative efforts, efficient communication, and continual innovation contributed to the development of the Mandala databases that manage taxonomic nomenclature, literature, illustrations, and over 122,000 specimens resulting from extensive worldwide expeditions and loans from over 200 collections. Yet concerns persist over ensuring data quality and addressing the long term maintenance of the data and database structure when the project terminates. This new collaborative paradigm is the future if we are to address taxonomic challenges posed by large, widespread groups in reasonable timeframes.

Digitized insect specimen access: new digitization tools

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In practise the overall efficiency of databasing the inventory of traditional entomological collections depends on two factors: suitable software and management measures to ensure the highest possible data quality already in the input process. Lessons learned from the development of the specimen-based database BIODAT, from the DORSA- (Digitized Orthoptera Specimen Access) project and preliminary results of the DIG-(Digitization of key Insect groups at ZFMK) project, which is especially designed to develop a 'good practise', recommend: (1) a lockstep programme for data-basing, (2) split record function, (3) visualisation of geo-referenced locations/sites during data entry, (4) semi-automatic/automatic data transformation from original format into alternative ones, (5) semiautomatic data transfer of taxo- and geo-referenced information units. Current activities deal with the introduction of semantic feedback mechanisms into the practise of data-basing entomological collections.

Three extraordinary genera of Dermestidae (Coleoptera) & their phylogenetic significance

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Three new genera of Dermestidae are described. One, the second known member of the subfamily Orphilinae, is represented by three species in eastern and southern Australia, two of which are associated with a conglobate and probably myrmecophilous type of larva. Another, from extreme northwestern Australia, is a member of the Dermestinae and shares features with both *Dermestes* and the Marioutini. The third genus from Malaysia and Indonesia appears to be intermediate between *Thyloias* and the Trinodinae. A cladistic analysis is performed on a data set including 19 genera of Dermestidae, representing all subfamilies and tribes, plus outgroups from Eucinetidae, Derodontidae, Nosodenidae, Endecatomidae and Bostrichidae, and about 100 characters, a third of which are based on immatures. The resulting cladograms are compared with recent studies on dermestid phylogeny.